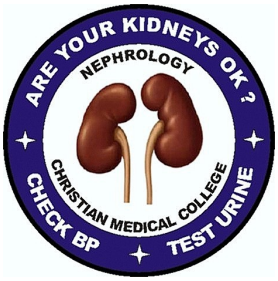
BODY COMPOSITION AND LEPTIN IN RENAL TRANSPLANTATION

A dissertation submitted to the Tamil Nadu Dr. M.G.R. Medical University in partial fulfillment of the University regulations for the award of

D.M. (Branch – III) (Nephrology)



Department of Nephrology

Christian Medical College, Vellore

BONAFIDE CERTIFICATE

This is to certify that the work presented in this dissertation titled "**BODY COMPOSITION AND LEPTIN IN RENAL TRANSPLANTATION**" done towards fulfillment of the requirements of the **Tamil Nadu Dr. M.G.R. Medical University, Chennai for the D.M. (Branch–III) (Nephrology)** exams to be conducted in Aug 2008, is a bonafide work of the candidate **Dr. VENKATA RAMANA RAJU S.,** Senior Post graduate student in the Department of Nephrology, Christian Medical College, Vellore under my guidance and supervision. This dissertation has not been submitted, fully or in part to any other board or University.

Guide & Head of Dept.:

Dr. George T. John M.D., D.M.,MNAMS, FRACP., FRCP,
Professor and Head,
Department of Nephrology,
Christian Medical College,
Vellore – 632004

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Contents

ABBREVIATIONS

DEXA – Dual Emission X ray Absorptiometry

MF-BIA – Multi Frequency Bioelectric Impedance Analysis

FM – Fat Mass

FFM – Fat Free Mass

BMI – Body Mass Index

BSA – body surface area

SFT – Skin Fold Thickness

BMC – Bone Mineral Content

BMD – Bone Mineral Density

DGF – Delayed Graft Function

CyA – Cyclosporine A

List of Tables:

List of Figures:

ABSTRACT

Aim: To assess the changes in body composition and leptin in the early post transplant period and to validate anthropometry in renal allograft recipients.

Patients and Methods: Consecutive renal allograft recipients were assessed prospectively at baseline, 3 & 6 months post-transplant by anthropometry, DEXA and serum leptin levels.

Results: 62 recipients (M:F=3:1, mean age 33.4±11.2 years), had a mean weight of 52±10.1, 56.8±9.3 and 57.7±9.6 kg at baseline, 3 & 6 months, respectively ($p<0.01$). The mean body fat at baseline, 3 & 6 months were 11.1±5.7, 13.9±5.7 and 14.5±6.2 kg respectively ($p<0.001$). The fat increment in arms, legs and trunk were 37.4%, 31.6% and 36.6% respectively. Skin fold thickness correlated well with fat (measured by DEXA) at all times (ICC=0.9, $p<0.001$). There was a 3.8% increase in lean body mass by 6 months ($p<0.01$) predominantly in legs. Anthropometry underestimated lean body mass. Total bone mineral content decreased from 1.92±0.34 to 1.85±0.31 kg ($p=0.001$) by 6 months, with significant reductions in spine (6.7%). The mean leptin levels at baseline, 3 and 6 months were 5.3±7.9, 6.0±8.4 and 15.4±17.4ng/ml respectively ($p<0.05$). At all times, leptin levels positively correlated with total body and regional fat content ($p<0.01$).

Conclusions: Post renal transplant, patients gain significant amount of weight, mostly due to accumulation of the fat, especially around arms & trunk .There is overall decrease in bone mineral content, predominantly in the spine. Anthropometric measurements for fat assessment are a reasonable substitute for DXA. Leptin levels correlate with body fat content.

INTRODUCTION

Weight gain is common after renal transplantation, influenced by improved appetite and a reversal of the uremic state. Renal transplant patients are at risk for increased weight, centripetal obesity and muscle atrophy because of their long-term glucocorticoid requirements. Such changes in body composition are associated with an increased risk of cardiovascular complications, which is a major cause of morbidity and mortality in renal transplantation since infective complications have decreased with newer immunosuppressive medication and therapeutic drug level monitoring. Body composition data might provide insight in to the relation with outcome, survival and post transplant complications and it might affect approaches to nutritional therapy and to therapy in the field of physical activity.[1]

Various techniques are available to measure body composition such as isotope dilution, anthropometry, dual energy X-ray absorptiometry (DEXA) and multi-frequency bioelectric impedance analysis (MF-BIA). Anthropometry comprises of series of noninvasive & inexpensive method of estimating body composition. However, it is operator dependant. Traditional methods of estimating total body fat rely on assumption that the fat distribution and bone mineral content are constant. However, in the patients undergoing renal transplantation rapid changes in body composition occur, with variations in fat distribution and bone mineral content.[2]

Dual-energy x-ray absorptiometry (DEXA) has been shown to be an effective measure of body composition, has been considered a valid and reliable reference measure, and has advantage of showing a three-dimensional model of body composition. However, it is expensive and available at limited centres.[3]

Marked central adiposity is one of the main characteristics of metabolic syndrome and previously adipose tissue was traditionally considered as energy storage organ but currently it is considered as endocrine organ secreting bioactive peptides called “adipokines” which has autocrine, paracrine and endocrine functions. Leptin being one such hormone regulate caloric intake and energy balance.

Data from India is limited with respect to body composition in the Post Transplant follow up period and the reliability of the anthropometry as compared to DEXA, which is expensive, and the role of leptin in the adiposity in renal transplant population.

REVIEW OF LITERATURE

Human Body Composition:

The major component of the human body is water. The protein and fat components are relatively small, with the remainder being primarily bone and minerals.

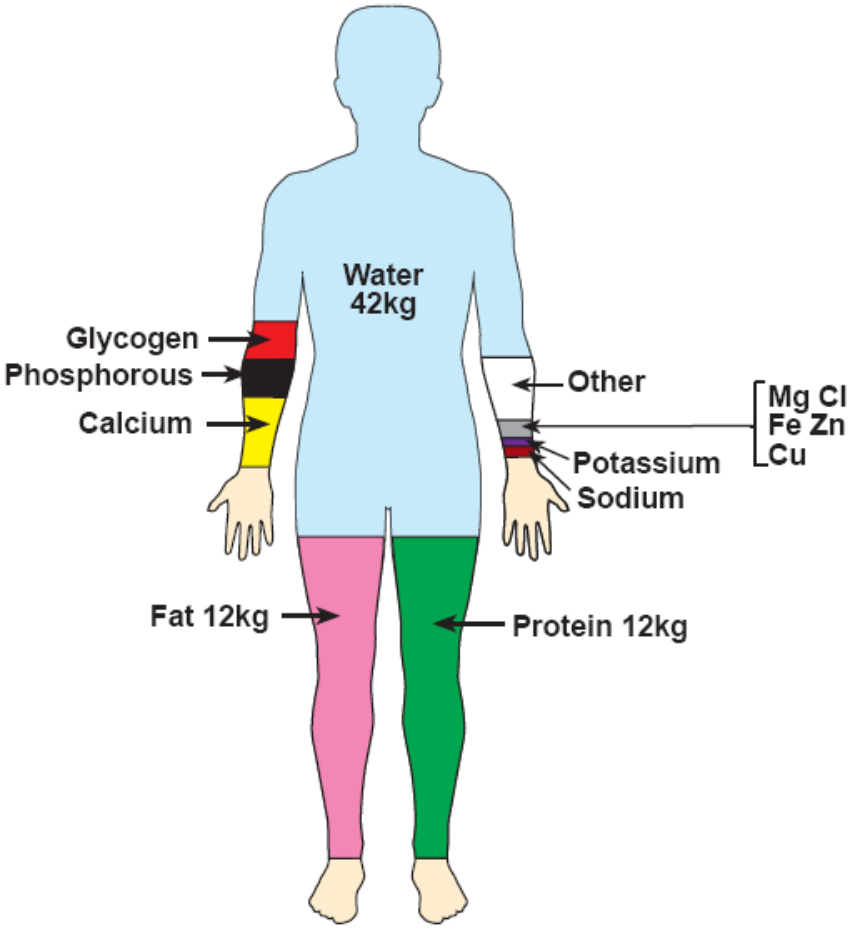


Figure 1. Human Body Composition

The non-fat component of body composition is termed fat free mass (FFM) and exists primarily as the chief structural and functional component of the human body. The FFM compartment consists in proportions of water (72%), protein (21%) and bone minerals (7%).

The fat compartment of the body is termed fat mass (FM) and will vary considerably between individuals in terms of absolute amount. Fat mass consists of 20% water and 80% adipose tissue and can, in obese persons be the largest component of the body.

Fat percentage:

Table 1. Fat percentages for various body compositions

Subject	*Normal*	*Overweight*	*Obese*
Adult Male	4-25%	25-30%	>30%
Adult Female	12-29%	29-35%	>35%

Comparison between lean and obese man:

Table 2. Body composition in lean and obese individuals

Content	*Lean man (70 kg)*	*Obese man (100 kg)*
Water	60%	47%
Protein	17%	13%
Fat	17%	35%
Remainder	6%	5%

There is a small amount of body protein available for energy, in the labile amino acid pool and muscle proteins during catabolism, (when the body is starving).

Carbohydrate is stored in the body typically as glycogen in the liver and in muscle and can vary between individuals ranging from approximately 500g in normal individuals to over a kilogram in trained athletes. Values also vary depending on body size and previous carbohydrate ingestion.

Elements:

The Mass of elements in a 70-kg person and the Volume of the purified elements are shown in the table 3[4].

Table 3. Volume and mass of elements in an average human

Element	**Mass(kg)**	**Volume(L)**	**Element**	**Mass(kg)**	**Volume(L)**
oxygen	43	37	potassium	.140	.429
carbon	16	7.08	sulphur	.140	.676
hydrogen	7.0	8.6	sodium	.100	.103
nitrogen	1.8	2.05	chlorine	.095	.063
calcium	1.0	0.645	magnesium	.019	.010
phosphorus	0.78	0.429	iron	.004	.005

Measurement of Body Composition:

Body composition measurements have been used to study how lean body mass (muscle, bone, etc.) and body fat change during health and disease, and have been used as a research tool in studying a wide variety of conditions, including metabolic and neuromuscular disorders, obesity, and the effects of aging. Some usefulness has also been reported in differentiating people who are large due to high lean tissue mass from those with high fat mass. Finally, there has been some research into the use of body composition calculations to make risk assessments regarding certain diseases and to assess the effects of exercise and nutritional programs in some patient populations.

The exact measurement of body composition is not possible outside the examination of a person's body after death. Given that limitation, the methods that are used to estimate body composition utilize predictive mathematical modeling along with body composition data collected with various techniques. All such techniques will present some variation and error based upon the underlying assumptions made while creating the formulae and their applicability on different populations.

Body composition methods are based upon a "compartment" model of the body, separating the body's components into two, three, or four compartments. Most methods only calculate two compartments, which are lean body mass (bone, muscle, tendons, etc.) and fat mass. Dual x-ray absorptiometry (DEXA), also calculates bone mass, a third compartment separate from lean body mass. Some models can be combined to add a fourth compartment, body water content. Several methods of measuring body

composition have been used over the past decade, including skin-fold measurement, hydrostatic weighing, bioelectrical impedance, dual x-ray absorptiometry (DEXA), and air displacement.

Anthropometry:

Skin-fold measurements, also called anthropometry, use a set of hand-held calipers exerting a constant known pressure to measure the thickness of various skin-folds, a portion of the skin pinched away from the body surface, at standard locations around the body. Either 3 or 7 skin-fold sites are used. Percentage body fat is calculated using the sum of the measurements. This technique is based upon the assumption that the thickness of subcutaneous fat reflects a constant proportion of total body fat. Several problems with this method are that the person making the measurements must be adequately skilled, and the calipers themselves must be accurately calibrated and have constant pressure. Finally, the more obese a person is the more difficult it is to "pinch" a skin-fold correctly, requiring a high level of technical skill to obtain accurate measurement.

Anthropometric Techniques:

Skin-fold thickness (SFT):- A skin-fold thickness (SFT) is defined as a measure of the double thickness of the epidermis, underlying fascia and subcutaneous adipose tissue. There are two main assumptions in determining total body fat from skin-folds:

1. That there is a constant relationship between total body fat and subcutaneous fat at the sites measured. The equation of Siri (1961) uses a two-compartment model, such that the human body

consists of fat mass (FM) and fat-free mass (FFM) and assumes that the density of the two compartments is constant between individuals at 0.90 g/cm3 for FM and 1.10 g/cm3 for FFM.

2. That the density of FFM is constant.

Collection of skin-fold thickness measurements (SFT) - Skin-fold measurements also assume that subcutaneous fat is a reliable indicator of total body fat and that skin-fold compressibility remains constant. Durnin and Womersley (1974) validated the sum of four SFT (biceps, triceps, subscapular and suprailiac) against densitometry and devised sex and age dependent population-based linear regression equations to estimate total body density. All SFT measurements should be taken by the same trained member of staff from identical positions on each subject, following the World Health Organization WHO 1987, anthropometric guidelines. Harpenden skin-fold calipers is used, with the subject in a standing position.

The four sites were as follows (Durnin and Womersley 1974):

1. ***Triceps***: A mark is made at the mid-upper arm, midline of the posterior aspect of the arm over the triceps muscle, measured with the elbow bent at 90º, used for identifying the biceps and triceps SFT. During the measurement, the arm should be hanging freely by the side, palms inwards towards the thighs.
2. ***Biceps***: Measured midline of the anterior aspect of the arm, over the biceps muscle, mid-point on the arm as described above.
3. ***Subscapular:*** Found just below and lateral to the bottom tip of the scapula, measured in a 45º angle. Subjects stand with their arm relaxed by their side. The scapula is palpated with the fingertips to find the bottom of the bone and the SFT is then measured in the natural crease. Subject's shoulders are relaxed.
4. ***Suprailiac (waist):*** Found 1 cm above the anterior superior iliac spine (top of the hipbone) in the mid-axillary line (waistline).Measured horizontally with the subject breathing gently.

To take the measurement, the skin is gripped about 1cm above the selected site and the calipers applied below this site, the grip is removed and the measurement noted to the nearest 0.2mm. The calipers are then removed. This is repeated for 3 successive measurements, with the mean value calculated.

Body density and percentage body fat is calculated using the equations of Durnin and Womersley (1974), for each side of the body, using the following equations:

$$\textbf{Density (g/cm}^3\textbf{) = c – m (log } \Delta S\textbf{)}$$

Where: D = Density, c and m = standard age and sex-specific coefficients

ΔS = Sum of all four skin-fold measurements (mm).

Once density is calculated, the Siri (1961) equation is used to estimate percentage body fat: **Fat (%) =**

$$\textbf{[(4.95 / D) – 4.5] x 100}$$

Where: D = Density, 4.95 and 4.5 are the constants calculated by Siri (1961) using the assumptions on the density of FM and FFM[5]

Lean body mass is calculated by the following James Equation[6]

$$\textbf{Lean Body Weight (men) = (1.10 x Weight(kg)) - 128 (Weight}^2\textbf{/(100 x Height(m))}^2\textbf{)}$$

$$\textbf{Lean Body Weight (women) = (1.07 x Weight(kg)) - 148 (Weight}^2\textbf{/ (100 x Height(m))}^2$$

Hydrostatic weighing:

Hydrostatic weighing, also known as underwater weighing, involves weighing a person when dry, then placing them into a tub or pool of water and measuring how much they weigh when totally submerged under water when the air in their lungs has been exhaled. Alternatively, the weight of the water displaced from the container being used may also be utilized. Using the knowledge that bone and muscle are more dense than water and

will sink, as well as the fact that fat is less dense than water and floats, the difference between the dry weight measurement and the submerged weight provides information which than can be used to estimate fat mass.

Limitations of this method include complaints about it being uncomfortable and cumbersome, variability in the ability of patients to fully exhale, variation in measurement due to movement of the subject, and the aversion to submersion of some patients. Additionally, the assumption that lean body mass density is constant is not accurate. Athletic individuals frequently have higher bone and muscle density, while older people frequently have lower bone density, leading to inaccurate calculations depending upon the equations used.

Bioelectrical impedance:

Bioelectrical impedance is based upon the theory that different compartments conduct electricity better than others. For instance, fat acts as an electrical insulator and does not conduct electricity well, while muscle conducts electricity quite well. This method involves using two metal surfaces with a small electrical potential between them. When the subject comes in contact with the plates simultaneously, electrical current flows through the subject. The resistance to the current caused by the patient's body is then used to calculate body composition data.

Limitations of this method are a higher standard error when compare to hydrostatic weighing and skin-fold measurement and dependency upon several variables which may be difficult to calculate accurately.

Dual Energy X-ray absorptiometry (DEXA):

Whole body dual energy x-ray absorptiometry (DEXA) is the only currently available measurement body composition that uses a three-compartment model. Lean body mass, fat mass, and bone mass are all estimated with this technique. This method is based on the principle that x-ray radiation is absorbed differently by body minerals. The more minerals contained in a material, the higher the absorption. Thus bone, which has a high mineral content, will absorb the radiation at a much higher rate than fat, which has a low mineral content.

This method uses two different low level x-ray beams, that when passed through the body, are absorbed at varying rates by the different compartments. This differential absorption of the x-rays is calculated to derive estimates of the three compartments. The radiation exposure when using this method has been reported to be very low. Limitations of this method include its inability to accurately measure body composition in extremely obese patients.

DEXA technology:

A typical DEXA instrument consists of a padded table on which the patient lies and a movable C-arm with an X-ray tube below the patient and a detector above the patient. The X-ray tube generates photon beams of two different energy levels, thus the term

"dual-energy." A collimator below the table limits the scatter of the photons and directs them toward the area of interest. The difference in attenuation (reduction in intensity) of the two photon beams as they pass through body tissue of variable composition distinguishes bone from soft tissue and allows quantification of BMD. Denser and thicker tissue contains more electrons and allows fewer photons to pass through to the detector. A computer with specially designed proprietary software designed by each manufacturer completes the DXA "system."

Radiation exposure to the patient is very small, usually of a similar magnitude to daily background radiation. Radiation scatter beyond the edge of the DXA table is negligible. No shielding of the technologist or the room is necessary. As a safety precaution, the technologist should not sit within three feet of the table edge while the patient is being scanned.

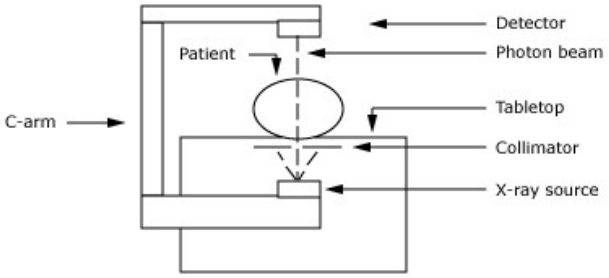


Figure 2. The DEXA apparatus

Plethysmography:

The use of body plethysmography, also know as air displacement plethysmography, is a technique that has been used for many years in the diagnosis and management of respiratory diseases. Only recently has this technology been applied to the measurement of body composition. Technically similar to the hydrostatic method but using air displacement instead of water displacement, this technique is relatively new. This method

involves a patient entering a sealed container with a known volume of air. As the patient breathes through a tube connected to the air outside the device, the air displaced from the interior of the container is measured. This volume of air, in conjunction with body weight is used to calculate body composition.

Limitations of this method are its limited availability, and variability in measurements due to hydration status, variation in atmospheric pressure affecting air density, and body temperature of the subject.

Body fat distribution:

The total body fat is distributed between visceral and nonvisceral (subcutaneous) compartments. Gonadal steroids play a major role in the distribution of body fat. At the onset of puberty, men become more muscular and have less fat, whereas women increase their body fat relative to their muscle mass. These differences persist throughout life and are reflected in the typical male and female fat distribution.

With age, both gonadal steroid and growth hormone secretion decline, which may explain the rise in visceral fat with age in men. In women, higher serum testosterone concentrations are usually associated with increased visceral fat. Thus, the decline in growth hormone and the loss of estrogen at the time of the menopause may explain the relatively rapid increase in visceral fat in postmenopausal women.

The distribution of body fat is important clinically. Visceral central adiposity is associated with a greater risk of metabolic and cardiovascular disorders including insulin resistance, type 2 diabetes mellitus, hypertension, and coronary heart disease [7-8]

Obesity and Renal Transplantation:

Weight gain is common after renal transplantation, which is influenced by improved appetite and a reversal of the uremic state. However, at least in the early post transplant period, the increase in body weight is mainly due to an increase in body fat mass. This phenomenon may be partly due to relatively high doses of steroids in the early period after renal transplantation, possibly mediated by their inhibiting effect on lipid peroxidation, but also appears to be related to physical inactivity.

Obesity is a major health issue in the Western world and now also in developing countries. Obesity also contributes to post transplantation cardiovascular morbidity, including NODAT[9], which is a major cause of death in patients with a functioning transplant. It is also associated with DGF in the case of cadaver kidney transplants, and the exact cause is not known.

Friedman et al[10] investigated the prevalence, demographics, and trends in obesity at the time of kidney transplantation from a transplant database that included all kidney transplantations since 1987. Body mass indices (BMIs) at the time of transplantation were stratified by demographic categories and year and compared with trends of obesity in the general population. They found that the majority (60%) of patients at the time of transplantation were overweight or obese; between 1987 and 2001, the proportion of obese transplant recipients increased by 116%.

Koolman et al[11]. observed that the post transplantation weight gain is, at least until 6 months after transplantation, predominantly due to an increase in fat mass and the increase in fat mass was already evident within 3 months after transplantation. Although extremity fat mass also increased, the changes in truncal fat mass were most pronounced.

Obesity increases the risk of surgery in general because patients are susceptible to delayed wound healing, difficulty in exposure of the operative site, and increased incidences of ventral hernias and deep-vein thromboses. However, in patients undergoing organ transplantation, there is an additional risk because these patients are on immunosuppression therapy, leading to increased risk for all types of infections, in particular, wound infections, and also delayed wound healing. Patients on sirolimus therapy have an additional risk for wound infections, delayed healing, and incisional hernias. Therefore, obesity confers an additional risk to patients undergoing kidney transplantation.

There is a curious association of obesity and delayed graft function (DGF) which is defined as the need for dialysis therapy in the first week after kidney transplantation. DGF is important for a number of reasons; there is an increased risk for chronic rejection and decreased graft survival, and it can be difficult to diagnose acute cellular rejection in patients who have prolonged DGF. DGF also leads to increased hospitalization and increased monitoring of the transplanted organ by means of noninvasive ultrasounds and graft biopsies. These issues lead to increased cost and risk for complications. The overall incidence of DGF is approximately 40%; however, with the use of perfusion pumps and induction therapy with antibodies, the incidence of DGF has been Reduced.4 Data from the USRDS including more than 51,000 patients receiving a kidney transplant found that a BMI greater than 36 kg/m2 was a major risk factor for DGF.

Howard et al[12] compared 3 groups of patients (BMI < 25 kg/m2, BMI between 25 and 30 kg/m2, and BMI >35 kg/m2). They found that incidences of DGF were 26.7%, 31.0%, and 36%, respectively, which was not statistically significant.

Series from St. Barnabas Medical Center, Livingston, NJ, found that the incidence of DGF was 24% versus 13% in patients with a BMI greater than 25 kg/m2 versus those with a BMI less than 25 kg/m2, respectively.[13]

Cardiovascular Morbidity after Kidney Transplantation

After kidney transplantation, patients experience increased cardiovascular morbidity and mortality caused by obesity, hypertension, post transplantation hyperlipidemia, and post transplantation diabetes mellitus (NODAT) as a result of steroids and calcineurin inhibitor medications. Enormous efforts are being directed to reduce cardiovascular morbidity by using steroid-reduction or steroid- withdrawal protocols. Some of the newer medications, such as sirolimus, also cause a greatly increased risk for hyperlipidemia, whereas steroid withdrawal should be balanced against a small, but definitive, risk for increased graft rejection.

Locsey et al[14] studied dyslipidemia and obesity in 137 patients. Although only 21.89% of patients had BMIs greater than 25.1 kg/m2 on dialysis therapy, after transplantation the proportion increased to 36.49%. In the group of patients treated with only cyclosporine A (CyA), the incidence of hyperlipidemia and hypertension was significantly less than in those administered a combination of steroids and CyA or steroids, CyA, and azathioprine. There was a definitive relationship between obesity and hyperlipidemia. They emphasized the importance of systematic control of lipid levels, diet with adequate carbohydrate and lipid content, and the necessity of avoiding obesity by selecting the optimal immunosuppressive treatment.

Baum et al[15] prospectively analyzed 506 renal transplant recipients between 1983 and 1998. In all transplant recipients, coronary artery disease was the most common cause of death, and African-American transplant recipients had the shortest graft survival and greatest percentage of deaths. At 1 year post transplantation, 39% of African-American transplant recipients were obese (BMI > 30 kg/m2), and odds ratios for NODAT were 3.5 and 5 times greater in nonwhite and obese transplant recipients, respectively. Multiple regression analysis confirmed the predominant independent effect of African-American race on weight gain; however, hypercholesterolemia was independent of race or ethnicity and was predicted by CyA treatment and NODAT.

Obesity and New onset Diabetes after transplantation (NODAT):

Kasiske et al[16] analyzed data from the USRDS with regard to 11,659 Medicare beneficiaries who received their first kidney transplant in 1996 to 2000. Cumulative incidences of NODAT were 9.1%, 16.0%, and 24.0% at 3, 12, and 36 months post transplantation, respectively. Risk factors for NODAT included age,

African-American race, Hispanic ethnicity, male donor, increasing HLA mismatches, hepatitis C virus infection, BMI of 30 kg/m2 or greater, and use of tacrolimus as the initial maintenance immunosuppressive medication.

Obesity and Long-Term Outcomes after Kidney Transplantation

Meier-Kriesche et al[17] retrospectively analyzed 51,927 primary adult renal transplantations registered in the USRDS. They categorized BMI values into 11 subgroups: less than 18 kg/m2, from 18 to 36 kg/m2 at 2-unit increments, and greater than 36 kg/m2. Primary study endpoints were graft and patient survival. Secondary study endpoints were death-censored graft survival, chronic allograft failure, DGF, and acute rejection. Using a variety of statistical methods, they found that extremes of very high and very low BMIs were associated with significantly worse patient and graft survival, but not with acute rejection. Elevated BMI also was associated with an increased risk for DGF, whereas lower BMI was significantly protective.

Yamamoto et al,[18] from Albany Medical Center, analyzed patient and graft survival in recipients of paired kidneys with similar preservation technique and surgical personnel. Between June1992 and August 1999, a total of 28 kidneys were transplanted into 1 obese and 1 nonobese recipient. DGF (7.1% versus 10.7%), acute rejection (39.3% versus 35.7%), and graft survival at 1 year were similar in the obese and nonobese recipient groups. Patient survival was similar at 1, 3, and 5 years in both groups. However, a trend toward decreased medium-term graft survival, which reached significance at 5 years, was observed in the obese group. Furthermore, mean serum creatinine level at 1 year was greater in the obese group (2.0 mg/dL [177 μmol/L]) compared with the nonobese group (1.4 mg/dL [124μmol/L]).

Meier-Kriesche et al[13], from St Barnabas Medical Center, Livingston, NJ, studied the effect of a BMI greater than 25 kg/m2 on long term renal transplantation outcome in 405 patients from 1990 to 1997. BMI greater than 25 kg/m2 were isolated as an independent risk factor for both decreased graft survival and patient survival (relative risk, 2.0 for each). Cadaveric donor status, acute rejection, and use of azathioprine versus mycophenolate mofetil were the only other significant risk factors.

Modlin et al[19], from the Cleveland Clinic, compared 127 obese(BMI >30 kg/m2) patients with a matched nonobese control group (BMI < 27 kg/m2) of 127 transplant recipients with similar demographics. There were no significant differences between groups according to donor source, recipient race or sex, re-transplantation, transplant percentage of reactive antibodies, cause of renal failure, or hypertension. Significantly more obese patients had a pre-transplantation history of angina or myocardial infarction. Mean follow-up was approximately 58.9 ± 40 months. Nonobese patients showed a significantly greater patient survival rate (89% versus 67%) at 5 years and only approximately half the number of deaths (25 versus 46 deaths) during the study. However, there were no significant differences between groups in DGF, acute rejection, chronic rejection, length of hospital stay, surgical blood loss, or mean serum creatinine level up to 5 years. Obese patients experienced a significantly greater incidence of NODAT (12% versus 2%).

Johnson et al[20] evaluated the effect of obesity on renal transplantation in a single Australian center between 1994 and 2000. Of 493 patients who underwent transplantation, 59 patients (12%) were obese (BMI > 30 kg/m2). Obese patients were more likely to experience superficial wound breakdown, complete wound dehiscence, and wound infections. There were no significant differences between the 2 groups with respect to surgery duration, postoperative complications, hospitalization, DGF, or acute rejection episodes. Five-year actuarial survival rates were similar between the 2 groups with respect to graft and patient survival.

Howard et al[21,] from the University of Florida College of Medicine, reviewed patients receiving a kidney transplant between 1990 and 1999 and grouped according to BMI. Group 1 had a BMI less than 25 kg/m2 (n =457); group 2, BMI of 25 or greater and less than 30 kg/m2 (n = 278); and group 3, BMI of 35kg/m2 or greater (n = 98). Cadaveric graft survival rates at 2 years were 85% for group 1, 88% for group 2, and 85% for group 3. Cadaveric patient survival rates at 2 years were 92% for group 1, 91% for group 2, and 94% for group 3. There were no differences in technical losses or post transplantation wound complications. However, group 3 patients had a greater incidence of NODAT. They concluded that obese transplant recipients have outcomes similar to nonobese patients.

Merion et al[22], from the University of Michigan Medical School, examined the influence of preexisting obesity (weight >120% of ideal body weight) on outcome after renal transplantation. Among 263 CyA-treated recipients of renal allografts, 223 patients (85%) were nonobese and 40 patients (15%) were obese before transplantation. Obese and nonobese transplant recipients were similar with regard to demographics, incidence of diabetes, and pre-transplantation serum cholesterol levels. There was a significantly greater incidence of wound infections in obese transplant recipients; other complications occurred with a similar incidence. There was no difference in incidence or number of rejection episodes; patient survival rates were 93% for nonobese patients at 3 years and 90.5%, which was not significant, for obese patients. Graft survival also was similar between the groups.

Drafts et al[23] reviewed their experience with living donor and cadaver transplantation in the current decade, focusing specifically on the impact of obesity on transplant outcome. Preoperative BMI was calculated for all adult renal transplant recipients between 1990 and 1995 and used to classify patients as nonobese, moderately obese, or morbidly obese. The effect of degree of obesity on early and late outcomes after renal transplantation was examined. Three hundred thirty-three transplant recipients had a pre-transplantation BMI less than 30 kg/m2 (normal or mild obesity), 68 patients had a BMI of 30 to 40 kg/m2 (moderate obesity), and 7 patients had a BMI greater than 40 kg/m2 (morbid obesity). There was no correlation between obesity and other demographic factors. Wound infections and DGF occurred more commonly in moderately and morbidly obese patients than in other cadaver donor recipients. Obese patients gained more weight after surgery and were administered lower CyA doses per kilogram. However, there was no significant correlation between obesity and graft survival for either cadaver or living donor transplants.

Treatment of Obesity after Kidney Transplantation:

Obesity continues to be a problem after successful kidney transplantation; however, this has not received as much attention as obesity at the time of listing for a kidney transplant.

Dietary Control

Lopes et al[24], from Spain, examined the nutritional status of 23 renal transplant recipients with a BMI greater than 27 kg/m2, hyperlipidemia, and stable renal function before and after 6 months of dietary intervention with the American Heart Association Step One pattern. After the dietary intervention, lipid profiles improved in all patients, with a decrease in mean total cholesterol level (237 versus 224 mg/dL [6.13 versus 5.79 mmol/L];(P <0.05), which was greater in men. Also, LDL cholesterol level was reduced in male patients (156 versus 136 mg/dL [4.03 versus3.52 mmol/L];(P<0.05), whereas in women, LDL cholesterol levels remained unaltered. HDL cholesterol and triglyceride values were not affected in men or women by the dietary treatment. The reduction in serum cholesterol level correlated inversely with initial triceps skin-fold value and was lower in patients with a BMI greater than 30 kg/m2 (5.7% versus 2.8%; P =NS). They concluded that obesity and hyperlipidemia after renal transplantation may be improved by dietary intervention.

Patel[25], from The Royal Hospitals, England, examined the effect of early intensive dietary intervention and follow-up on weight gains in new renal transplant recipients. Group A was studied prospectively, and group B was studied retrospectively during 1 year post transplantation. Group A consisted of 11 patients (9 men, 2 women) who underwent transplantation consecutively during 2 months, with a mean age of 39 years. Group B consisted of 22 patients (14 men, 8 women) who had undergone transplantation consecutively 4 years before the study, with a mean age of 40 years. Both groups had functioning grafts and similar triple immunosuppressive therapy (prednisolone, CyA, and azathioprine). Group A received intensive individualized dietary advice in stages, with regular follow-up for the first 4 months post transplantation, whereas group B did not receive dietary advice. Patel found that patients in group A showed significantly lower weight gains compared with group B at 4 months and 1 year. At 1 year, group A had a mean weight gain of 5.5 kg/patient compared with 11.8 kg/patient in group B. Intensive dietary advice is an effective tool to prevent weight gain in kidney transplant recipients. This may take the form of a specialized diet, such as the American Heart Association Step One, or another specific diet plan suggested by a qualified dietician.

Surgery

Bariatric surgery before or after kidney transplantation has been reported in the form of case reports. Marterre et al [26], from the University of Cincinnati Medical Center, performed Roux-en-Y gastric bypass in 3 morbidly obese (200% to 260% of ideal body weight) patients 6 to 8 years after kidney transplantation. Both patients who developed NODAT had complete resolution within 9 months after bypass surgery. On average, patients required 3 fewer hypertension medications after surgery; 2 of the 3 patients were off medication, with significant improvement in hyperlipidemia.

Steroid Withdrawal

Modern immunosuppressive regimens have succeeded in reducing the risk for acute rejection; however, long-term dangers of cardiovascular disease and other post transplantation complications of immunosuppressive therapy persist. Long-term use of steroids is linked with a well known pattern of side effects on skin, bone, and the cardiovascular system in renal transplant recipients. Hyperlipidemia, which is increased in patients administered steroids, is one of the most important risk factors for cardiovascular diseases[27]. Steroid withdrawal has been shown to definitely reduce cholesterol and LDL levels in recipients of solid-organ transplants, and there is a trend toward reduction of obesity[28]. Steroid withdrawal will lead to a reduction in cardiovascular morbidity; however, this has to be balanced against the small increased risk for acute rejection.

Psychotherapeutic Programs

Weight gain and its prevention is a concern in a majority of the population. However, weight gain among hemodialysis patients and transplant recipients is particularly worrisome because studies have suggested a direct correlation between obesity, quality of life, and compliance behavior. Obesity has been addressed successfully by using behavioral modification, a combination of nutritional education and supportive counseling among patients with diabetes. Melin et al[29] associated the most successful outcomes with continuous feedback and positive re-enforcement from the counselor, along with realistic goal setting.

Other programs have successfully combined physical activity regimens, counseling, and nutritional advice among general population cohorts.

Leptin:

Leptin was discovered as a result of studies of ob/ob mice, a strain of hyperphagic obese mice that were known to lose weight when their circulation was attached to normal mice (parabiosis) [30]. Subsequent studies revealed that ob/ob mice had a mutation that results in inability to produce a protein, first called the ob protein and later leptin that regulates food intake[31]. In addition to being very obese, these mice grew poorly and had infertility due to gonadal hypofunction. Administration of leptin to these animals resulted in a marked decrease in food intake, weight loss, and improved growth [32]. Other obese mice, diabetic (db/db) mice and fatty (Zucker) rats have genetic defects in the leptin receptor [33]. Db mice are phenotypically identical to ob mice.

In humans, the leptin gene is located on chromosome 7q32 and consists of three exons and two introns that span 20 kilobases (kb) of DNA. The mouse and human ob genes have 84 percent homology. The gene codes for a secreted protein of 167 amino acids.

Mechanism of Action of Leptin :

Leptin is a member of the cytokine family, and its receptor is a member of the gp130 group of cytokine receptors. There are at least five forms of the leptin receptor [34]. The most widely distributed is the short form of the receptor, which is present in most tissues and may serve to transport leptin into the brain. The long form of the receptor is located in areas where leptin is thought to act, including hypothalamic nuclei. There may also be a circulating form of the leptin receptor that binds leptin.

The signaling system on the intracellular portion of the leptin receptor belongs to the janus kinase signal transduction and translation system (JAK STAT). It is the Stat-3 form of the STAT system that is thought to carry out the intracellular signaling [35]. A counter-regulatory system that inhibits leptin and cytokine action exists in the suppressors of cytokine signaling, which occur in at least three different forms. Parenteral administration of leptin increases mRNA levels for one form of this suppressor in the hypothalamus, liver, and small intestine, which may explain the occurrence of resistance to the action of leptin [36].

Circulating factors that bind leptin might also contribute to leptin resistance. In one study, C-reactive protein was identified as a circulating factor that binds to leptin, impairs its signaling, and attenuates its physiologic effects (in cultured cells and an ob/ob mouse model) [37]. In addition, physiologic concentrations of leptin stimulated C-reactive protein expression in vitro.

Food intake:

Food intake is reduced by systemic leptin administration in normal-weight experimental animals, but the response decreases as the animals become obese. However, when leptin is injected into the ventricular system of the brain of obese animals, they remain responsive [38]. Since leptin is transported across the blood-brain barrier to act within the brain, the processes controlling the entrance of leptin into the brain are pivotal determinants of its action on food intake [39].

Leptin decreases food intake in several ways. It decreases the content of neuropeptide Y (NPY) mRNA [(40-41)] and it increases the content of proopiomelanocortin (POMC) mRNA in the arcuate nucleus of the hypothalamus[42]. NPY is one of the most potent stimulators of food intake [40], and alpha-melanocyte-stimulating hormone (alpha-MSH), produced by cleavage of POMC, inhibits food intake. Thus, leptin acts in the arcuate nucleus to reduce food intake in two ways, decreasing NPY and increasing alpha-MSH.

The importance of these effects is illustrated by the following observations. Leptin-deficient ob mice that are also deficient in neuropeptide Y are less obese than ob mice in which neuropeptide Y production is normal [43]. Furthermore, mice lacking POMC-derived peptides are obese and lose weight when treated with alpha-MSH[44.]

Prothrombotic effect:

Leptin also may have a prothrombotic effect. Leptin-deficient mice form an occlusive thrombus more slowly than wild-type mice, a change reversed by leptin replacement [45].

This effect appears to be mediated through the platelet leptin receptor.

Bone formation:

Data on the effect of leptin on bone metabolism are conflicting. In obese mouse models deficient in leptin (ob/ob) or its receptor (db/db), bone formation and bone mass have been reported to be increased [46], decreased [47], or variable depending upon the bone site[48.] It appears that leptin's effects on energy balance and bone occur at the level of the hypothalamus, although the pathways appear to be distinct [46].

Data in the human are also inconclusive, as observational studies have reported both a positive [48, 49, 50] and negative [51, 52, 53, 54] association between serum leptin concentrations and bone density. However, in a study of exogenous leptin administration in women with hypothalamic amenorrhea (who are leptin-deficient[55], an Increase In Markers Of Bone Formation Was Observed[56].

Biology of Leptin:

Leptin is produced primarily in fat cells, and also in the placenta and probably in the stomach. Large fat cells produce more leptin than small ones and serum leptin concentrations are highly correlated with body fat content in newborn infants, children, and adults [57, 58]. Leptin mRNA and secretion by adipocytes declines rapidly during starvation. These processes are stimulated by insulin, glucocorticoids and tumor necrosis factor-alpha, another product of adipocytes. These observations suggest that leptin signals the brain about the quantity of stored fat.

Leptin production in humans:

Features of leptin production in humans other than the correlation with body fat content include:

1. **Serum leptin concentrations increase with progressive obesity.** The concentrations are higher in women than in men, for any measure of obesity [59, 60], and they decrease with age in both women and men [61]. Pregnant women have higher serum leptin concentrations than nonpregnant women [62]. Breastfeeding may reduce the risk of child obesity [63], and leptin could possibly play a role, as it is produced in the breast and is present in milk [64, 65]. Serum leptin concentrations increase during childhood, with the highest concentrations in children who gain the most weight; higher serum leptin concentrations are associated with an earlier onset of puberty [66]. The potential importance of leptin in this setting is illustrated by the observation that mice and rats deficient in leptin fail to undergo pubertal development, while the administration of leptin to such animals results in pubertal onset [67].

Serum leptin concentrations are similar in black and white children of similar body composition [68, 69]. The concentrations are similar in normal subjects and patients with type 2 diabetes mellitus of the same weight; chronic endogenous hyperinsulinemia does not increase leptin secretion, although infusion of insulin and glucose for two days does [70].

2. There is a **diurnal rhythm of serum leptin concentrations**, the values being 20 to 40 percent higher in the middle of the night as compared with daytime [70,71]. The peak shifts in parallel with shifts in the timing of meals [72]. In human beings, plasma leptin is related to blood pressure levels in both normotensive [73] and hypertensive subjects [74, 75].
3. **Leptin production is strongly influenced by nutritional state**. Overeating increases serum leptin concentrations by nearly 40 percent within 12 hours, long before any changes in body fat stores. Conversely, in both normal-weight and obese subjects, fasting reduces serum leptin concentrations by 60 to 70 percent in 48 hours [76].

CLINICAL STUDIES

Genetic disorders:

Leptin deficiency:

Congenital leptin deficiency due to a mutation in the leptin gene, produces massive obesity, similar to that seen in rodent models that lack leptin or leptin receptor [77, 78]. Early-onset obesity and profound hyperphagia are characteristic of these individuals, as are hyperinsulinemia and advanced

bone age [79, 80, 81]. Hypogonadotrophic hypogonadism occurs in some patients [80].

Serum leptin levels were significantly lower in heterozygous members of these families than would be expected from their percent body fat. However, these heterozygotes had normal thyroid function, appropriate gonadotropin levels and normal secondary sexual characteristics, including menstrual cycles. Leptin deficiency also reduces TSH pulsatility.

Food intake was dramatically reduced when three leptin deficient children were treated with leptin [79]. Pubertal development was seen in one child, and serum concentrations of free thyroxine and TSH increased into the normal range.

In a second study, leptin replacement therapy for 18 months in three morbidly obese homozygous leptin-deficient adults resulted in dramatic weight loss, and resolution of type 2 diabetes and hypogonadism [81]. There was an initial decrease in food intake (energy intake) which reached a nadir at four to six months, but which then gradually returned towards baseline, but this was offset by a progressive increase in physical activity.

Leptin receptor deficiency:

Human obesity resulting from a mutation in the leptin receptor (LEPR) has been described [82, 83]. In one report, 8 of 300 subjects (3 percent) with severe, early-onset obesity had nonsense or missense LEPR mutations (six

probands were from consanguineous families). In addition to severe obesity and hyperphagia, other characteristics of affected patients included:

1. Alterations in immune function (decrease in the absolute CD4+ T-cell count with compensatory increase in the CD19+ B-cell count).
2. Normal linear growth, but reduced adult height as adults (due to lack of pubertal growth spurt).
3. Delayed puberty due to hypogonadotropic hypogonadism.
4. Increased serum leptin concentrations (consistent with their elevated fat mass, but not disproportionately increased, suggesting that serum leptin levels are not a useful marker for LEPR deficiency).
5. Less severe clinical features when compared to patients with congenital leptin deficiency (less hyperphagia, lower BMI, and lower percentage body fat).

Obesity:

Most obese people, however, are not leptin deficient, and serum leptin concentrations are directly related to their amount of body fat. In several surveys of obese subjects, no mutations in the leptin gene were detected [84, 85]. Given the high serum leptin concentrations and apparent leptin resistance in obese subjects [59, 60] little response to exogenous leptin might be expected. However, in a study of the effect of recombinant leptin (0.01, 0.03, 0.1, or 0.3 mg/kg per day) or placebo in normal-weight subjects for four weeks and obese subjects for 24 weeks, there was a very modest dose-dependent decrease in weight in both groups [86]. After weight loss, leptin administration

prevents the decline in metabolic rate and circulating concentrations of thyroid hormone [67].

Hypothalamic amenorrhea:

Leptin administration for the relative leptin deficiency in women with functional hypothalamic amenorrhea (due to weight loss, excessive exercise, or an eating disorder) [55, 56] may improve function of the reproductive axis (increased serum LH concentrations and pulsatility) as well as the thyroid and growth hormone axes [87, 88].

Lipodystrophy:

Leptin-replacement therapy is effective in patients with lipodystrophy and leptin deficiency.

Immune Function:

Leptin deficient individuals have a decrease in CD4 cells and reduced T-cell production [79] that are consistent with their higher rates of childhood infection [80] Leptin replacement leads to a switch from secretion of predominantly Th2 cytokines to Th1 cytokines [79].

AIM

1. To assess the changes in body composition and leptin in the early post transplant period in renal allograft recipients
2. To validate anthropometric measurements using DEXA in renal allograft recipients.

PATIENTS AND METHODS

Design and Location:

This prospective study was conducted at the Department of Nephrology, Christian Medical College (CMC), Vellore.

Inclusion Criteria:

a) Renal allograft recipients with 6 months of follow-up.

b) DEXA scans and serum samples for Leptin analysis available at baseline, 3rd and 6 months.

Exclusion Criteria:

a) Renal allograft recipients with less than 6 months of follow-up.

b) DEXA scans or serum samples for Leptin not available at any of the point in the study period.

Duration:

Patients who underwent renal transplantation at CMC, Vellore between May 2006 to Feb 2007 were included and followed up till August 2007

Methodology:

Renal allograft recipients, on inclusion, underwent evaluation at baseline, 3 months and at 6 months after their transplant operation. All patients underwent anthropometric evaluation, whole body DEXA scans and Serum leptin level estimation at these time points. Clinical details were noted from the patient follow up.

Measurements:

1. **Anthropometry:**

    i. **Height and Weight** – Height and weight of patients were measured on height and weighing scales, standardized to the rules of Department of Legal Metrology of the Government of India. Anthropometric calculations were based on the following formulae:

1. Lean Body Weight in Kg = **LBW=[(1.10-(0.03 X sex] X (weight)-{[128 + (sex X 20)] X [(weight/height)^2]**
2. Body mass index in Kg/m^2 **=BMI = Weight in Kg / (Height in ms)^2**

ii. **Skin Fold Thickness** (SFT): SFT were measured using Harpenden's calipers and estimation of body fat done by method of Durnin and Wormesley as detailed below. The four sites were as follows (Durnin and Womersley 1974):

5. ***Triceps***: A mark is made at the mid-upper arm, midline of the posterior aspect of the arm over the triceps muscle, measured with the elbow bent at 90º, used for identifying the biceps and triceps SFT. During the measurement, the arm should be hanging freely by the side, palms inwards towards the thighs.
6. ***Biceps***: Measured midline of the anterior aspect of the arm, over the biceps muscle, mid-point on the arm as described above.
7. ***Subscapular:*** Found just below and lateral to the bottom tip of the scapula, measured in a 45º angle. Subjects stand with their arm relaxed by their side. The scapula is palpated with the fingertips to find the bottom of the bone and the SFT is then measured in the natural crease. Subject's shoulders are relaxed.
8. ***Suprailiac (waist):*** Found 1 cm above the anterior superior iliac spine (top of the hipbone) in the mid-axillary line (waistline).Measured horizontally with the subject breathing gently.

To take the measurement, the skin is gripped about 1cm above the selected site and the calipers applied below this site, the grip is removed and the measurement noted to the nearest 0.2mm. The calipers are then removed. This is repeated for 3 successive measurements, with the mean value calculated.

Body density and percentage body fat is calculated using the equations of Durnin and Womersley (1974), for each side of the body, using the following equations:

$$\textbf{Density (g/cm}^3\textbf{)} = c - m\ (\log \Delta S)$$

Where: D = Density, c and m = standard age and sex-specific coefficients

ΔS = Sum of all four skin-fold measurements (mm).

Once density is calculated, the Siri (1961) equation is used to estimate percentage body fat:

Fat (%) = [(4.95 / D) – 4.5] x 100

Where: D = Density, 4.95 and 4.5 are the constants calculated by Siri (1961) using the assumptions on the density of FM and FFM[5]

2. **DEXA:** Whole Body scan was performed using HOLOGIC DEXA apparatus for whole and regional body fat, bone mineral content and lean body mass.
3. **Serum Leptin:** Fasting serum samples for the Leptin were drawn and stored at -70º centigrade. All Serum samples for Leptin were analyzed at 6 months by Leptin (Sandwich) ELISA. (DRG Instruments GmbH, Germany).

Statistical Analysis:

1. **Measures of central tendency and distribution** – Mean ± standard deviations were used for normally distributed data and median & range (min – max) was used for skewed data to avoid the outlier effect.
2. An **intra-class correlation coefficient** (one way random) was calculated for the Anthropometry and DEXA measurements and their significance was determined. Scatter plots were used for graphical representation of the same.
3. Bias and precision were calculated based on the following definitions and graphically demonstrated using **Bland -Altman** analysis plots
    i. **Bias** = Mean difference (x – y)
    ii. **Precision** – 2 Std. deviation of Bias = 2 X S.D (x – y)

    x, y – variables.

RESULTS

Patient Profile: It is a prospective study from May 2006 to February 2007. 73 patients underwent renal allograft transplantation of which three patients had less than 6 months follow up period, 7 patients had not performed one of the 3 sets of DEXA scans and 1 patient was excluded from the study in view of moribund obesity. Statistical analysis was performed on 62 patients.

Demographic Data:

Among 62 patients, 45 were males (72.58%)

The mean age:

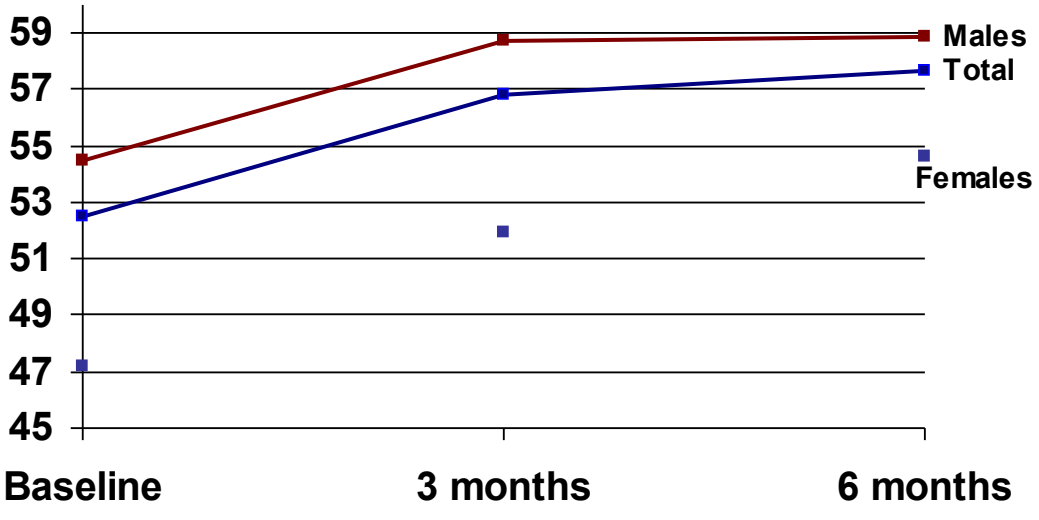
- Male-32.31±11.20 years
- Female-36.29±8.91 years

Body Weight:

Table 4. Temporal Profile of Weight

Weight (Kg)	Baseline (mean±SD)	3 months (mean±SD)	6 months (mean±SD)
Male (n=45)	54.51 ± 10.00	58.72 ± 9.07	58.97 ± 9.58
Female (n=17)	47.16 ± 8.91	51.94 ± 8.33	54.62 ± 9.18
Total (n=62)	52.49 ± 10.19	56.86 ± 9.32	57.78 ± 9.60

Figure 3. Temporal profile of body weight



Body Mass Index:

Nearly half of the study population (45.2%) were underweight (BMI <18.5) at the entry of study, which reduced to 21% by3 months and 17.7% by the end of 6 months the distribution of normal weight (BMI-18.5-24.9) and over weight(BMI- 25-29.9) category as shown in the table below

Table 5. Body Mass Index

BMI	baseline	3 months	6 months
<18.5	28(45.2%)	13(21.0%)	11(17.7%)
18.5-24.9	30(48.4%)	41(66.1%)	40(64.5%)
25-29.9	4(6.5%)	8(12.9%)	11(17.7%)

Figure 4. Temporal Profile of Body Mass Index

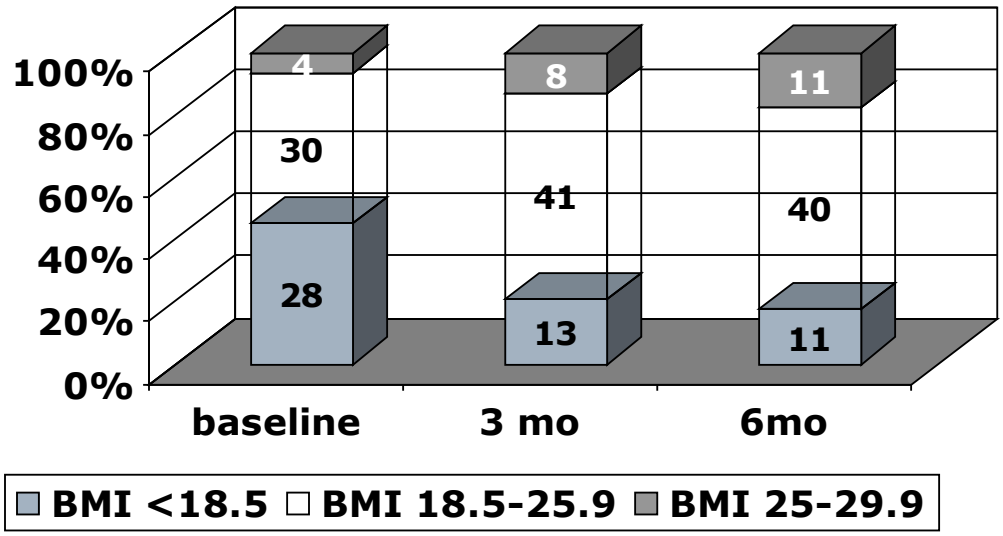


Table 6. Body Mass Index profile in Males and Females.

BMI	Gender	Baseline	3 months	6 months
<18.5	**Total**	**28(45.2%)**	**13(21.0%)**	**11(17.7%)**
	Male	20(44.4%)	9(20%)	9(20%)
	Female	8(47.0%)	4(23.5%)	2(11.8%)
18.5-24.9	**Total**	**30(48.8%)**	**41(66.6%)**	**40(64.5%)**
	Male	22(48.9%)	31(68.9%)	30(66.7%)
	Female	8(47.0%)	10(58.8%)	10(58.9%)
25-29.9	**Total**	**4(6.5%)**	**8(12.9%)**	**11(17.7%)**
	Male	3(6.7%)	5(11.1%)	6(13.3%)
	Female	1(6%)	3(17.7%)	5(29.3%)

DEXA Fat: There is statistically significant increase in the fat content (in gm) over a period of 6 months in all the sub regions of the body except the head fat. However the head fat gain significant in females (p=0.04). Over a period of 6 months, the arm fat

Increased by 37.4%, and increment was 378.7gm in males vs. 912.4gm in females, trunk fat increment was 36.6 %, with increment of 1753.8 gm in males vs. 2480.1 gm in females and leg fat increment was 889.4gms in males vs. 1716.4 gm in females with total increment of 31.6%.

Figure 5. Temproal profile of Total Body and regional fat

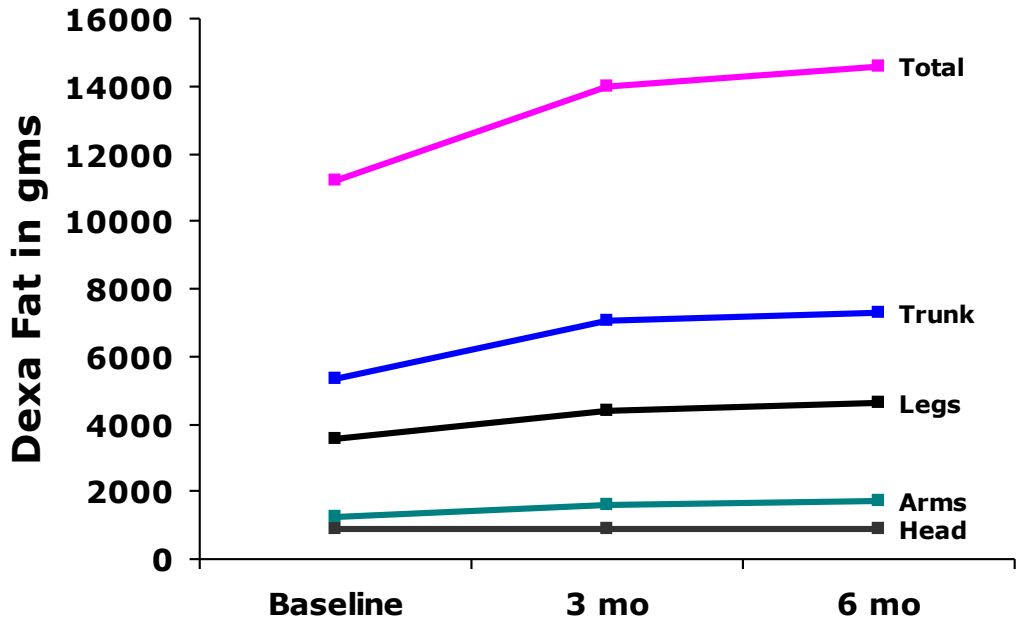


Table 7. Body fat composition changes with time

Fat (Gms)	Baseline (Mean±SD)	3 months (Mean±SD)	6 months (Mean±SD)	p
Head	861.17±104.45	888.79±108.53	881.49±148.81	0.21
Trunk	5336.13±3319.64	7057.28±3233.09	7291.84±3491.24	**<0.01**
Arms	1268.21±818.97	1626.80±868.43	1743.92±1015.75	**<0.01**
Legs	3524.28±1814.22	4394.70±1966.65	4640.43±2027.97	**<0.01**
Total body	11178.55±5787.04	13961.78±5761.53	14576.68±6205.24	**<0.01**

Table 8. Fat content for males and females with time

Fat (gm)	Sex	Baseline (mean±SD)	3 months (mean±SD)	6 months (mean±SD)	p
Head	**Male**	890.95±94.81	912.43±109.84	901.04±162.55	0.51
	Female	782.33±88.18	826.20±77.43	829.73±88.56	**0.04**
Trunk	**Male**	4873.81±3071.40	6580.21±3083.49	6627.65±3241.60	**<0.01**
	Female	6559.84±3725.51	8320.12±3371.99	9049.99±3612.14	**<0.01**
Arms	**Male**	1094.54±691.22	1419.91±727.71	1473.27±822.04	**<0.01**
	Female	1727.94±966.35	2174.44±989.90	2640.35±1150.60	**<0.01**
Legs	**Male**	3024.32±1528.18	3840.55±1814.80	3913.73±1619.11	**<0.01**
	Female	4847.68±1887.44	5861.58±1593.31	6564.07±1747.43	**<0.01**
Total	**Male**	10203.69±5318.91	12747.12±5315.62	12919.64±5369.05	**<0.01**
	Female	13759.05±6335.02	17177.05±5806.87	18962.96±6273.94	**<0.01**

Body fat in gm by Anthropometry & correlation with DEXA:

The body fat as derived by anthropometric measurements at 3 time points is given in the table below. The intra class coefficient between the fat measured by anthropometry and to that of DEXA were 0.91, 0.93 & 0.93 at baseline, 3 months and at 6 months ($p<0.01$) showing a very good agreement between anthropometric measurement of fat and by DEXA.

Table 9. Correlation between Anthropometry and DEXA fat measurements

Fat	Baseline (mean±SD)	3 mo (mean±SD)	6 mo (mean±SD)
Anthropometry (Kg)	**10.42±5.92**	**13.42±5.91**	**14.76±6.76**
DEXA (Kg)	**11.17±5.78**	**13.96±5.78**	**14.57±6.20**
ICC	**0.91**	**0.93**	**0.93**

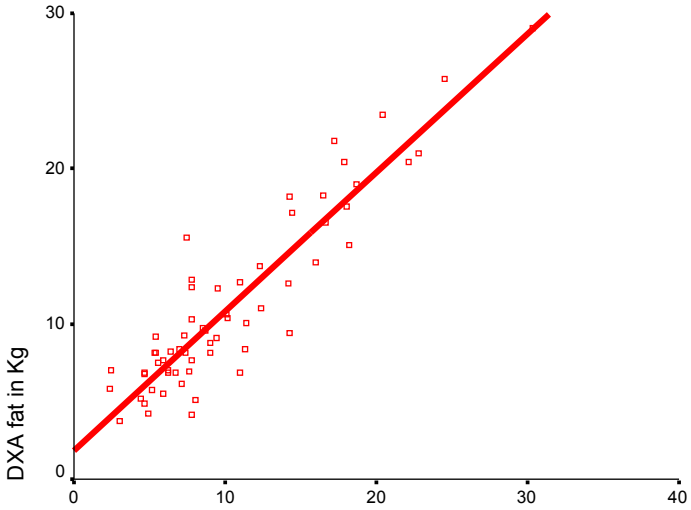


Figure ***ıts at baseline***

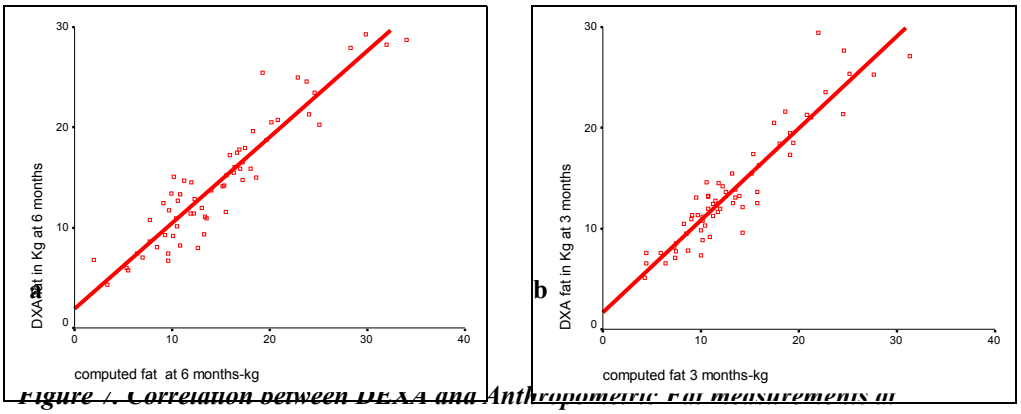


Figure 7. Correlation between DEXA and Anthropometric Fat measurements at

(a) 3months and (b) 6 months

Bland Altman Analysis:

Table 10. Bias and Precision Analysis of Anthropometric Fat measurement to DEXA fat measurement

Fat (Kg)	Bias (Mean difference)	Precision (SD)	Limits of agreement
Baseline	+0.751	2.14	-3.97 to 5.47
3 months	+0.541	2.08	-3.56 to 4.64
6 months	-0.186	2.40	-4.88 to 4.51

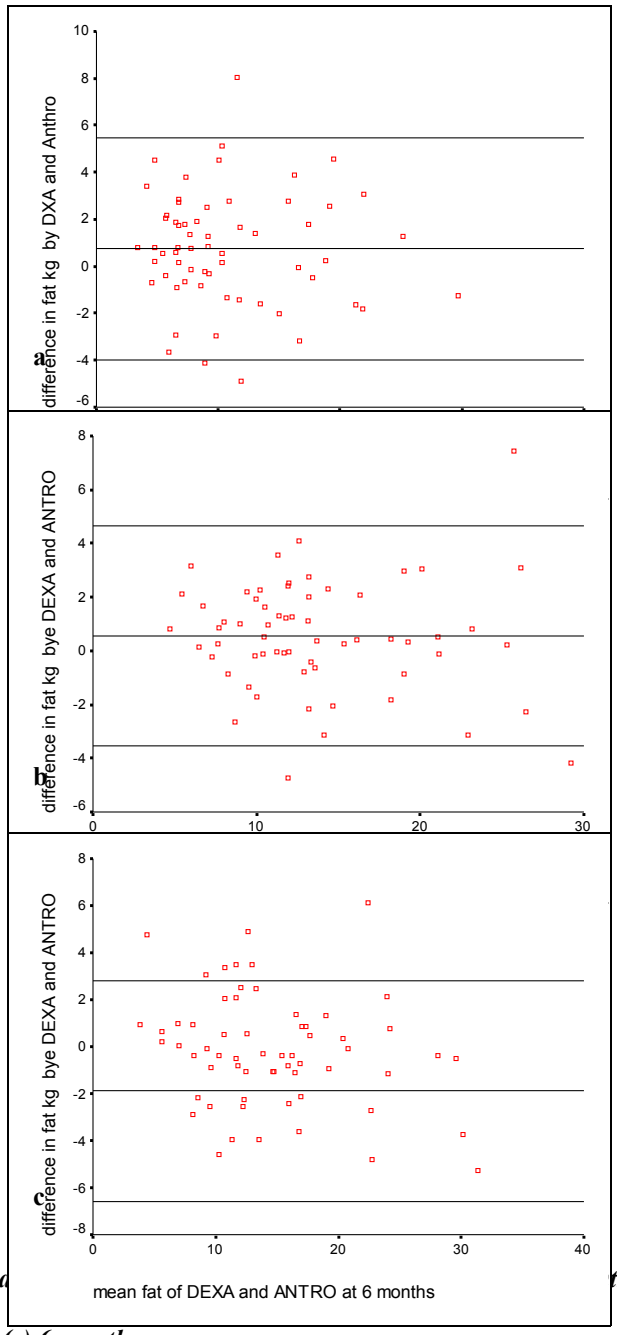


Figure 8 Bla… …try and DEXA at (a) baseline, (b) 3 months and (c) 6 months

The results of Bland Altman Analysis are given in the table above. The Limits of agreement show that the anthropometry may be 3.97 kg lower or 5.47 kg higher than DEXA measured at baseline, 3.56 kg & 4.88 kg lower or 4.64 kg & 4.51 kg higher at 3 months and 6 months respectively.

Fat Percentage:

Parallel to fat in gm, fat in percentage increased over time significantly except head ($p<0.01$) and between sex, it followed similar pattern of fat in gm.

Table 11. Temporal profile of total and regional fat percentages

Fat %	Baseline (Mean±SD)	3 months (Mean±SD)	6 months (Mean±SD)	p
Head	18.53±0.80	18.77±0.67	18.81±0.55	0.18
Trunk	20.65±9.30	24.93±8.34	25.34±8.91	**<0.01**
Arms	21.33±11.14	25.93±10.90	26.85±12.03	**<0.01**
Legs	21.90±9.37	24.78±8.62	25.57±9.62	**<0.01**
Total body	20.97±8.44	24.53±7.84	25.11±8.54	**<0.01**

Table 12. Temporal profile of total and regional fat percentages in males and females

Fat %	Sex	Baseline (Mean±SD)	3 months (Mean±SD)	6 months (Mean±SD)	p
Head	Male	18.58±.92	18.78±.74	18.78±.59	0.21
	Female	18.38±.33	18.74±.42	18.88±.42	**<0.01**
Trunk	Male	18.32±7.76	22.46±7.00	22.47±7.39	**<0.01**
	Female	26.81±10.56	31.47±8.25	32.94±8.24	**0.001**
Arms	Male	17.45±7.86	21.68±7.85	21.75±8.52	**<0.01**
	Female	31.59±12.20	37.21±9.86	40.36±9.23	**<0.01**

Legs	Male	17.91±6.11	21.07±6.24	21.02±6.14	**<0.01**
	Female	32.45±8.33	34.62±5.87	37.62±6.03	**<0.01**
Total fat	Male	18.17±6.42	21.55±6.00	21.68±6.32	**<0.01**
	Female	28.39±8.80	32.42±6.64	34.20±6.86	**0.001**

Fat	**Baseline (Mean±SD)**	**3 months (Mean±SD)**	**6 months (Mean±SD)**
Anthropometry fat (%)	18.99±8.54	23.10±8.22	24.93±9/30
DEXA fat (%)	20.97±8.44	24.53±7.84	25.11±8.54
ICC	0.88	0.92	0.91
p	<0.01	<0.01	<0.01

Body fat in percentage by Anthropometry & correlation with DEXA:

The body fat % as derived by anthropometric measurements at 3 time points is given in the table below. The intra class coefficient between the fat % measured by anthropometry and to that of DEXA were 0.88, 0.92, & 0.91 ($p<0.01$) at baseline, 3 months and 6 months showing a very good agreement between anthropometry and DEXA with respect to fat in percentage

Table 13. Correlation between Anthropometry and DEXA fat percentage measurements

Table 14. Bias and Precision analysis for fat percentage by Anthropometry and DEXA

Fat %		
Baselin		.09
3 montl		53
6 montl		76

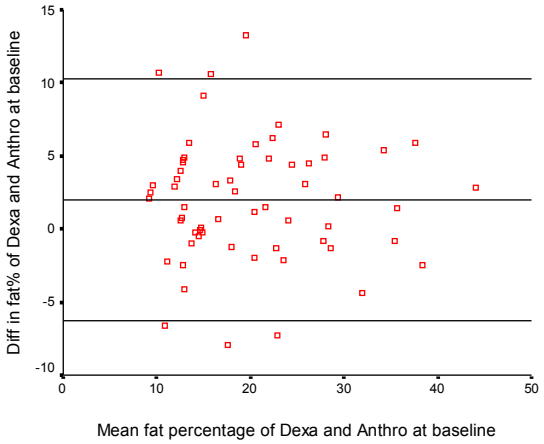
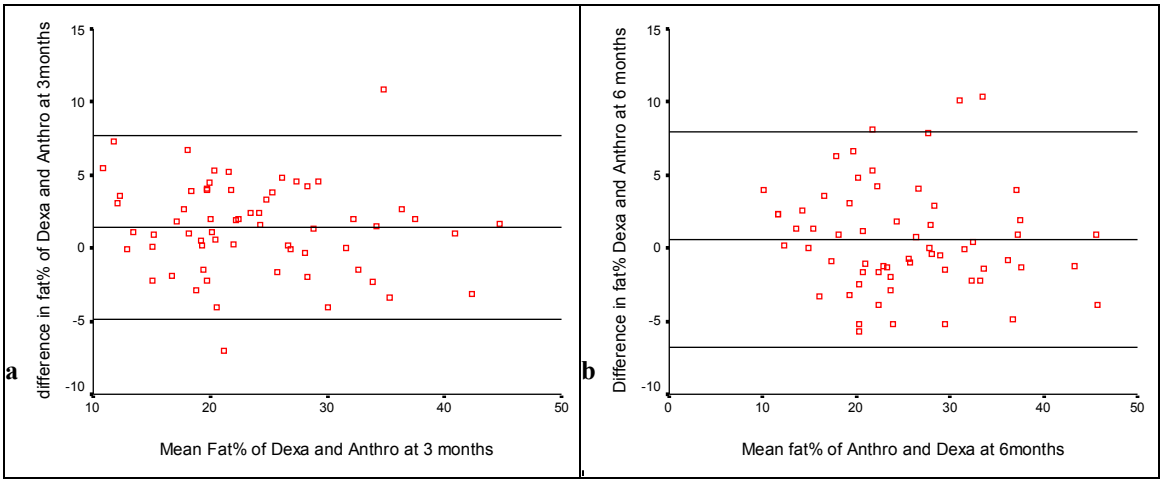


Figure 9. Bland Altman Plot for fat % by anthropometry and DEXA at baseline



The results of Bland Altman Analysis are given in the table above. The

Limits of agreement show that the anthropometry may be 6.13% lower or 10.09% higher than DEXA measured at baseline, 4.69% & 6.58% lower or 7.53% & 7.76% higher at 3 months and 6 months respectively.

Bone mineral content:

There is overall decrease in the bone mineral content in all the sub regions of the body, but significant in the regions of trunk (3.8%), spine (6.7%) and pelvis (6.9%) and the whole body (3.5%). Males tend to have more reduction in the bone mineral content compared to females.

Table 15. Temporal profile of Bone Mineral Content

BMC	Baseline	3 months	6 months	p

	(Mean±SD)	(Mean±SD)	(Mean±SD)	
Arms	271.94±55.68	264.87±60.13	265.26±60.39	0.07
Ribs	145.48±35.91	150.49±36.10	148.51±35.47	0.17
Trunk	468.35±91.39	454.42±90.39	450.17±90.00	**0.01**
Spine	147.37±25.63	138.76±25.68	137.34±24.68	**<0.01**
Pelvis	176.10±43.49	165.16±38.50	163.82±41.54	**<0.01**
Legs	693.68±148.94	688.26±144.03	686.06±138.21	0.39
Total body	1928.62±344.73	1873.87±314.13	1859.97±310.15	**0.01**

Table 16. Temporal profile of Bone mineral content in males and females

BMC	**Sex**	**Baseline (Mean±SD)**	**3 months (Mean±SD)**	**6 months (Mean±SD)**	**p**
Arms	Male	292.23±47.82	289.18±50.27	288.69±52.06	0.40
	Female	218.24±36.20	200.52±27.81	203.23±28.58	0.14
Ribs	Male	155.27±34.57	161.14±34.02	156.16±36.51	0.18
	Female	119.57±25.45	122.29±24.93	128.25±23.05	**0.03**
Trunk	Male	492.83±88.81	477.76±89.33	468.69±93.04	**0.01**
	Female	403.58±63.30	392.64±60.15	401.14±59.66	0.26
spine	Male	151.67±25.85	142.91±26.21	139.45±25.92	**<0.01**
	Female	135.99±21.85	127.79±21.19	131.76±20.72	0.07
Legs	Male	745.85±135.23	738.38±133.81	732.46±129.61	0.20
	Female	555.59±79.80	555.57±63.86	563.24±67.41	0.60
pelvis	Male	186.72±44.12	173.70±39.38	172.42±42.73	**<0.01**
	Female	148.00±26.49	142.55±25.26	141.07±28.29	0.15
Total	Male	2036.99±321.99	1973.56±295.01	1949.19±301.68	**0.02**
	Female	1641.74±217.92	1609.99±186.94	1633.79±186.57	**0.04**

Bone mineral density: The bone mineral density significantly decreased in all the sub regions of the body except pelvis. Males had significant reduction in the BMD ($p<0.01$) as compared to females except in pelvis.

Table 17.Temproal Profile of Bone Mineral Density

BMD (g/cm^2)	**Baseline (Mean±SD)**	**3 months (Mean±SD)**	**6 months (Mean±SD)**	**p**	**% loss**
Arms	1.46±0.14	1.45±0.13	1.44±0.13	**<0.01**	**1.36**

Ribs	1.28±0.17	1.26±0.15	1.24±0.15	**<0.01**	**3.12**
Spine	1.79±0.20	1.72±0.20	1.70±0.25	**<0.01**	**5.02**
Pelvis	0.96±0.12	0.95±0.12	0.94±0.12	0.14	2.08
Legs	2.23±0.27	2.18±0.25	2.15±0.24	**<0.01**	**3.58**
Total body	1.06±0.09	1.03±0.08	1.02±0.08	**<0.01**	**3.77**

Table 18. Temporal profile of bone mineral density in males and females

BMD (g/cm^2)	**Sex**	**Baseline**	**3 months**	**6 months**	**p**
Arms	Male	1.52±0.11	1.51±0.10	1.49±0.11	**<0.01**
	Female	1.31±0.07	1.30±0.07	1.29±0.06	0.46
Ribs	Male	1.32±0.16	1.30±0.15	1.27±0.16	**<0.01**
	Female	1.16±0.12	1.16±0.09	1.16±0.10	0.84
spine	Male	1.80±0.20	1.72±0.19	1.69±.21	**<0.01**
	Female	1.72±0.19	1.72±0.24	1.73±0.33	0.67
Legs	Male	2.32±0.24	2.26±0.24	2.22±0.23	**<0.01**
	Female	2.00±0.18	1.96±0.13	1.95±0.15	0.06
pelvis	Male	0.95±0.12	0.94±0.12	0.94±0.12	0.18
	Female	0.96±0.13	0.96±0.13	0.95±0.12	0.57
Total	Male	1.07±0.87	1.05±0.08	1.03±0.08	**<0.01**
	Female	1.02±0.90	1.00±0.07	0.99±0.80	**<0.01**

Lean body weight:

There was overall increase in the lean body weight ($p=0.01$) and legs in particular ($p<0.01$). Similar findings were seen between the sub category of males and females.

Table 19. Temporal profile of Lean Body Weight

Lean (gms)	**Baseline**	**3 months**	**6 months**	**p**
Arms	4156.00±1072.69	4255.84 ±993.89	4292.30 ±1030.18	0.14

Legs	11833.29±2752.61	12456.22±2357.50	12570.69±2310.53	**<0.01**
Trunk	19553.29 ±3700.51	19775.16 ±3910.36	20144.51 ±3203.20	0.18
Head	3324.97±400.27	3388.31 ±378.97	3396.11 ±420.17	0.04
Total body	38921.78 ±7391.14	40241.09±6440.98	40403.58 ±6694.60	**0.01**

Table 20. Temporal profile of Lean Body Weight in males and females

Lean (gms)	**Sex**	**Baseline**	**3 months**	**6 months**	**p**
Arms	Male	4631.58±851.84	4704.34±754.13	4722.97±840.99	0.55
	Female	2897.12±267.30	3068.62±372.62	3152.27±445.58	**0.03**
Legs	Male	12948.92±2308.44	13451.73±1915.95	13491.15±1915.95	**0.006**
	Female	8880.15 ±1210.74	9821.05±963.77	10134.18±935.31	**<0.01**
Trunk	Male	20860.46±3270.05	20853.09±3957.76	21285.56±2916.70	0.45
	Female	16093.12±2296.63	16921.81±1817.35	17124.11±1539.03	**0.02**
Head	Male	3450.30±354.81	3490.38±369.15	3504.16±415.52	0.31
	Female	2993.24±321.76	3118.14±256.78	3110.10±280.20	0.11
Total lean	Male	41905.93±6217.45	43003.22±5125.67	43001.65±5882.86	**0.04**
	Female	31022.55±3259.37	32929.58±2767.67	33526.35±2541.43	**<0.01**

Lean body weight (LBW) by Anthropometry & correlation with DEXA:

The lean body weight derived from weight of the individual by James equation at 3 time points are given in the table below. The James equation overestimates LBW by 12.28%, 12.97% and 13.82% at baseline, 3 months and 6 months respectively. The intra class coefficient between anthropometric measurement of lean body weight and DEXA were 0.88, 0.90 & 0.88 at baseline, 3 months and 6 months respectively ($p<0.01$), showing a good agreement between anthropometric lean and DEXA lean.

Table 21. Correlation between lean mass measurement by Anthropometry and DEXA

Lean body weig	6 months (Mean±SD)
Computed Lean	46.88 ± 6.68
DEXA lean bod	40.40 ± 6.69
ICC	0.88
p	<0.01

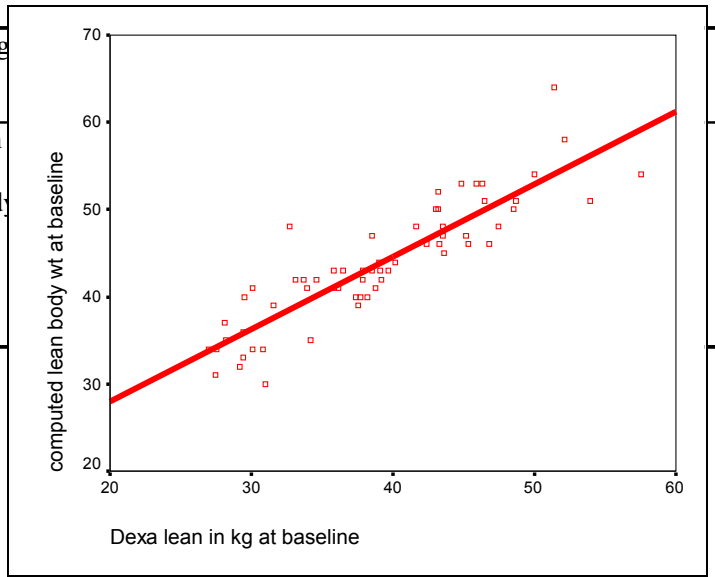
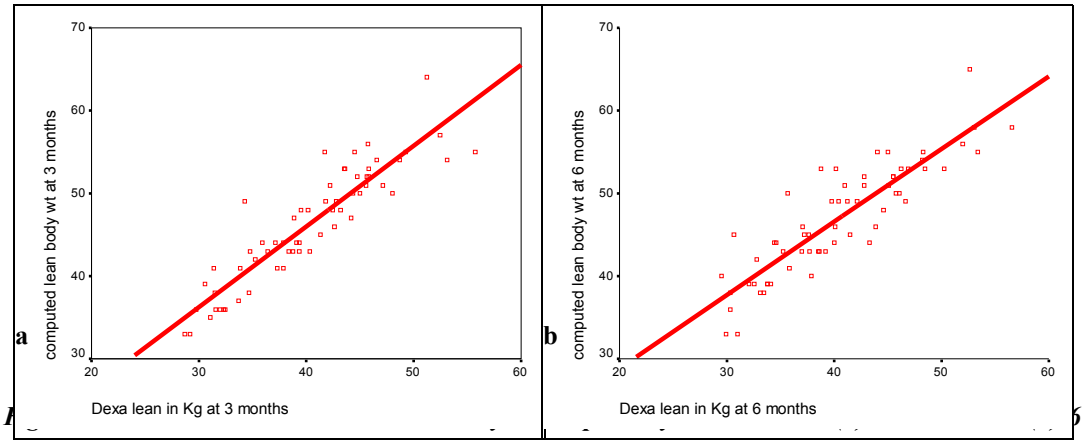


Figure 11. Correlation between Lean mass by Anthropometry and DEXA at basleine



months.

Bland Altman analysis: The results of Bland Altman Analysis are given in the table below. The Limits of agreement show that DEXA may be 11.60 kg lower or 2.04 kg higher than anthropometry measured at baseline, 0.41kg to 11.58kg lower at 3months and 0.12kg to 12.83 kg lower than anthropometric measurements at 6 months. Thus anthropometry overestimates lean body weight in Indian population.

Table 22. Bias and Precision analysis for Lean mass by Anthropometry and DEXA

Lean body (Kg)	Bias (Mean difference)	Precision (SD)	Limits of agreement
Baseline	-4.78	3.48	2.04 to -11.60
3 months	-6.00	2.85	-0.41 to -11.58
6 months	-6.48	3.24	-0.12 to -12.83

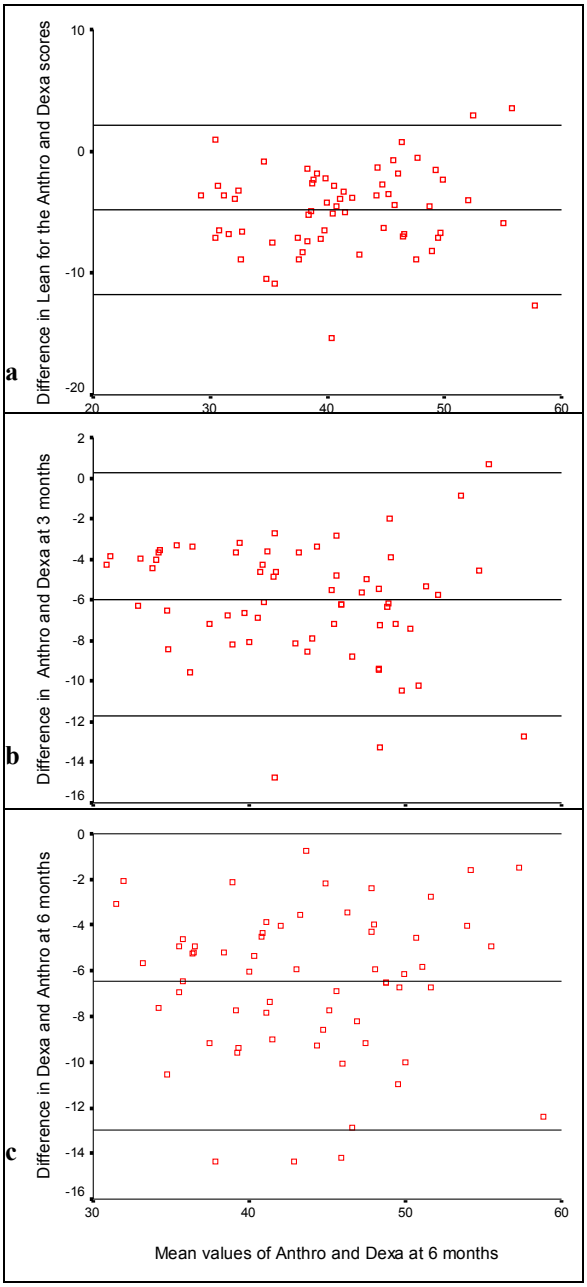


Figure 13. Bland Altman plots for lean mass by Anthropometry and DEXA at (a) baseline, (b) 3 months and (c) 6 months

Leptin:

The mean leptin levels increased from 5.28ng/ml at baseline to 6.09ng/ml at 3 months to 15.48ng/ml at 6 months. The females had higher level of leptin compared to males at all the points of time (8.5ng/ml vs 4.07 ng/ml at baseline, 10.99ng/ml vs. 4.23ng/ml at 3 months and 31.30ng/ml vs. 9.50ng/ml at 6 months.)

Table 23. Temporal Profile of Leptin levels

Leptin	**Baseline**	**3 months**	**6 months**
ng/ml	**ng/ml**	**ng/ml**	**ng/ml**
Male (n=45)	4.07±7.59	4.23±7.56	9.50±12.08
Female (n=17)	8.50±8.04	10.99±9.02	31.30±19.84
Total (n=62)	5.28±7.90	6.09±8.48	15.48±17.45

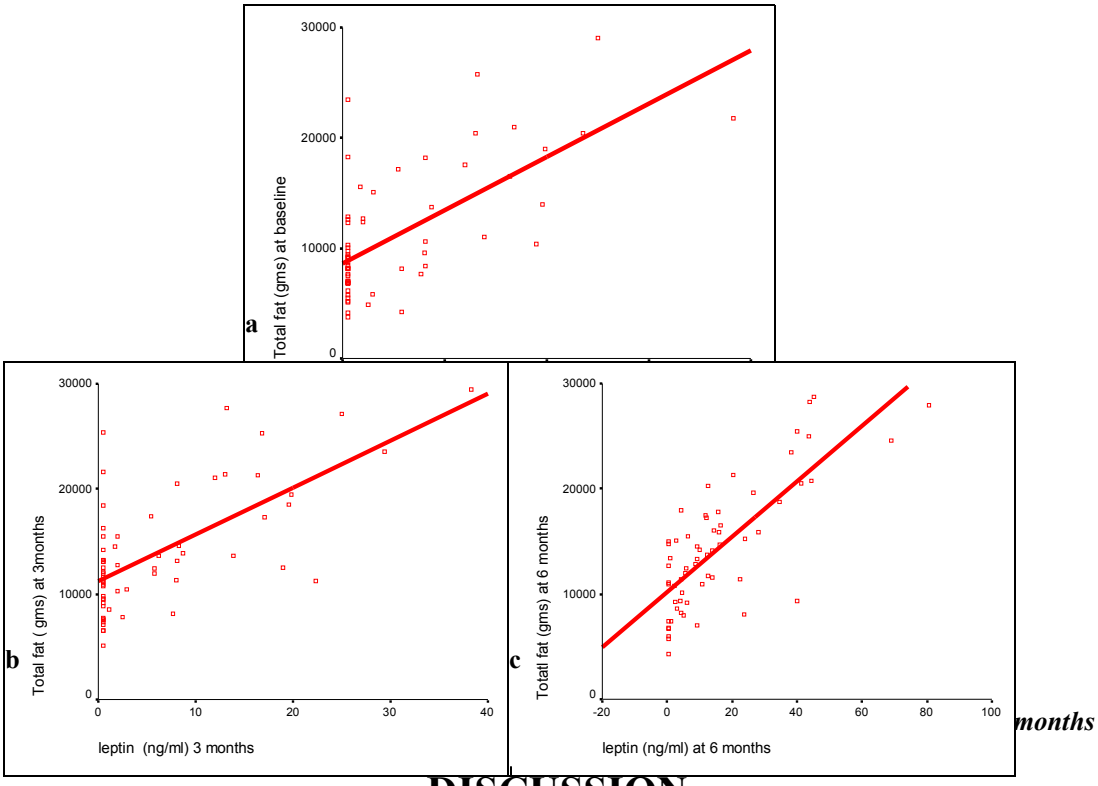
Correlation of leptin with body fat, bone mineral content (BMC), bone mineral density (BMD) and lean body mass:

The correlation of leptin with respect to fat in "gms" & percentage, bone mineral content and its density, lean body weight is given in the table below. Leptin has good correlation with respect to fat in gms (ICC being 0.65, 0.65 and 0.81 at baseline, 3 months and 6 months respectively) and fat percentage (ICC being 0.61, 0.65 and 0.77 at baseline, 3 months and 6 months respectively. Leptin has poor correlation with respect to bone mineral and lean body tissue.

Table 24. Correlation between Leptin and Fat, BMC, BMD and lean mass by DEXA

Leptin		**Total Fat (g)**	**Total Fat (%)**	**BMC (g)**	**BMD (g/cm^2)**	**Lean (g)**
baseline	r	**0.65**	**0.61**	-0.52	-0.05	-0.20
	P	**<0.01**	**<0.10**	0.68	0.66	0.87
3 months	r	**0.65**	**0.65**	-0.08	-0.03	-0.17
	P	**<0.01**	**<0.01**	0.49	0.80	0.16
6 months	r	**0.81**	**0.77**	-0.05	-0.03	-0.18

p	**<0.01**	**<0.01**	0.66	0.79	0.14



months

DISCUSSION

In our study, we assessed the change in body composition over a period of 6 months using DEXA with respect to body fat, bone mineral content, bone mineral density and lean body mass. In addition, agreement between measurements made by anthropometry and DEXA for the body fat and lean body mass was evaluated. Finally, the correlation of leptin with body fat, bone mineral and lean body mass was analyzed.

Weight Gain: There is progressive weight gain in both genders during the first 6 months. Nearly half of the study populations were underweight immediately after renal transplantation. However, 66% of patients had normal BMI at 3 and 6 months. Only 12.9% and 17.7% of patients were over weight at 3 and 6 months respectively. Females tend to gain weight more than males over a period of time and were more over weight.

In the study by Cofana et al [89], 38% of the patients were over weight and 16% were obese. Obesity was more prevalent in women (21% vs 13%). In our study however, only 12.9% of the patients were over weight and most often they were women (6.0% vs 6.7% at baseline, 17.7% vs 11.1% at 3 months and 29.3% vs 13.3% at 6 months). Males had attained their near maximal weight by 3 months, whereas, females continued to gain weight till 6 months

Fat Gain: There is significant increase in the total & regional fat over a period of 6 months except for the head fat. Patients tend to accumulate more fat in arms and trunk.

In the study by Kooman et al [90], weight gain observed in the first few months after renal transplantation is predominantly due to an increase in body fat mass, the changes in truncal fat mass being most pronounced. This was consistent with finings in our study.

DEXA has been validated for the assessment of the fat, however it is expensive. Skin fold thickness is a well-established, simple and inexpensive technique for determining body fat in normal adults. In the present study, we wanted to assess the validity of anthropometric measurements as compared to DEXA in renal allograft recipients. Anthropometry underestimated fat content at baseline and 3 months (mean bias of -0.75 and 0.55 kg respectively). However, it overestimated fat content at 6 months compared to DEXA (mean bias of -0.19Kg). These differences are negligible in the clinical situation concerned. However, anthropometry consistently underestimated fat % measured by DEXA (bias 4.69% to 10.09%).

In the study by Hart PD, et al [91]. who analyzed 34 renal transplantation patients at time of transplantation, and again after 3 and 6 months, they found the ICC between DEXA and skin fold thickness were 0.84, 0.78 & 0.85 respectively at ($p<0.01$). and in 34 control patients, the ICC was 0.95 ($p<0.01$) and skin fold measurements underestimated changes in the total body fat, especially those gaining substantial amounts of body fat.

In our study, the correlations were good both with fat and fat %. Even though there are some discrepancies between skin fold thickness and DEXA, they appear to be minimal and hence skin fold thickness can be used as reliable measure of body fat.

Bone mineral: Renal transplantation improves the metabolic environment and restores the glomerular filtration and renal production of 1,25-dihydroxy vitamin D3 which is impaired during chronic renal failure, in spite of this bone loss is a frequent and well-known complication in the first months of after renal transplantation particularly at trabecular bone sites [92, 93].

In our study, there is decrease in the bone mineral content of whole body in general and trunk, spine, pelvis in particular. Males tend to have more reduction. The bone mineral density however, reduced in all the sub regions except pelvis and maximal loss of BMD occurred in the spine. In the study by Fabio et al [93] involving 20 males, the bone mineral density reduced only in ribs and pelvis. Hober et al [94] showed that baseline total body and compartmental BMD was approximately 9% lower than in controls; during the 5-month follow-up, the BMD of the limbs remained unchanged, where as that of the trunk and head decreased.(3)

Kwan et al [95] found significant decrease of total body BMD (approximately 2.5%) at 3 months, followed by partial recovery at 6 months with significant bone loss also in the spine and femoral neck with no subsequent recovery.

Further more Cuteo-Manzano et al [96]. indicated male sex as one of the strongest predictive factors for low bone mass in long term renal transplantation

Lean Mass: Muscle wasting is well known consequences of kidney transplantation and it is related to glucocorticoid therapy. Mathieu et al [97]. found a decrease in lean mass and increase in trunk fat mass in kidney recipients treated with immunosuppressive monotherapy (CsA). Steiger et al [98] found a decrease of

approximately 10% in the limb and trunk lean mass in comparison with controls and, during the course of the follow-up, an increase in fat mass and limb lean mass in male patients; reduction in trunk lean mass observed during the first 2 months of follow-up and remained unchanged there after. Fabio et al [93] found no change in total and sub regional lean mass. Ham et al [99] did not find any relationship between body composition and different steroid dosages.

Contrary to the decrease in the lean mass in the above studies, in our study, there was increase in lean mass in general and legs in particular ($p < 0.01$). In addition, arm lean mass in females also increased significantly ($p = 0.03$). This is probably due to increased physical activity following renal transplantation. James's equation over estimated Lean body mass (mean bias -6.0 Kg)

Leptin: Leptin plays an important role in regulating appetite and energy expenditure and also functions in the neuroendocrine, hematopoietic, and immune systems, among others. Leptin may be involved in modulating bone mineralization. females tend to have more leptin as studied by Ostlund et al [100]. Agras et al [101] showed elevated leptin level is associated with increased bone mass at lumbar sites in renal transplant recipients, suggesting its bone-sparing effect.

Serum Leptin levels correlated with the percentage of body fat, trunk fat, lean body mass, serum creatinine, and urea. It was not related to nutritional status, BMD, or bone metabolism in kidney allograft recipients as seen in study by Malyaszko et al.[102] It is also not related to excretory graft function and immunosuppression as shown by Franciszek et al. [103]

In our study, the leptin increased over a period of time. Females had elevated leptin at all points of time. Leptin correlated with fat, but had no relationship to BMD or lean mass.

Conclusions

In renal allograft recipients,

1) There is significant weight gain after transplantation, more commonly in women and most often due to an increase in body fat (especially in arms and trunk)
2) The bone mineral content and density decreases with time, often in men and predominantly in the spine.
3) The lean body mass increases marginally with time, predominantly in the legs.
4) Skin fold thickness is a reliable and valid measure of body fat
5) James equation overestimates lean body mass measured by DEXA.
6) Serum leptin level increased over a period of time predominantly in females.
7) Serum leptin levels correlate well with fat content, but not with BMD or lean mass.

BIBLIOGRAPHY

1. Eugenie CH, Van Den Ham, et al. Body composition in Renal Transplant Patients: Bioimpedance analysis compared to Isotope Dilution, Dual energy X ray absorptiometry and anthropometry. J Am Soc Nephrol.: 1999, 10; 1067-1079.

2. Hart PD, Wilkie ME, et al. Dual energy x-ray absorptiometry versus skinfold measurement in the assessment of total fat in renal transplant recipients. Eur J Clin Nutr. 1993 May; 47(5):347-52.

3. Rodney G Bowden, Beth A Lanning et al . Comparison fo body composition measures to Dual Energy X ray Absorptiometry. JEP online 2005; 2.vol 8.

4. Emsley, John, *The Elements*, 3rd ed., Clarendon Press, Oxford, 1998.

5. National Kidney Foundation. K/DOQI guidelines 2000. appendix VII.

6. James WPT. Research on obesity. London. Her Majesty's Stationery Office. (ISBN 0-11-4500347). 1976.

7. Peiris AN, Sothman MS. et al. Adiposity, fat distribution, and cardiovascular risk. Ann Intern Med 1989; 110:867.

8. Lapidus L, Bengtsson C, et al. Distribution of adipose tissue and risk of cardiovascular disease and death: a 12 year follow-up of participants in the population study of women in Gothenburg, Sweden. BMJ 1984; 289:1257.

9. Jindal RM, Hjelmesaeth J: Impact and management of posttransplant diabetes. Transplantation 2000;70:58-63.

10. Friedman AN, Miskulin DC. et al:Demographics and trends in overweight and obesity inpatients at time of kidney transplantation. Am J Kidney Dis 2003; 41:480-487.

11. Kooman, Eugenie C. H. van den Ham Jeroen P. Et al. Post transplantation weight gain is predominantly due to an increase in body fat mass. Transplantation. 2000; 70(1):241-242, July 15.

12. Howard RJ, Thai VB et al: Obesity does not portent a bad outcome for kidney transplant recipients.Transplantation 2002; 73:53-55.

13. Meier-Kriesche H-U, Vaghela M, et al: The effect of body mass index on long-term renal allograft survival. Transplantation 1999; 68:1294-1297.

14. Locsey L, Asztzlos L, et al:The importance of obesity and hyperlipidemia in patients with renal transplants. Int Urol Nephrol 1998; 30:767-775.

15. Baum CL, Thielke K,Westin E, et al: Predictors of weight gain and cardiovascular risk in a cohort of racially diverse kidney transplant recipients. Appl Nutr Invest 2002;18:139-146.

16. Kasiske BL, Snyder JJ, et al: Diabetes mellitus after kidney transplantation in the United States.Am J Transplant 2003; 3:178-1185.

17. Meier-Kriesche H-U, Arndorfer JA, et al: The impact of body mass index on renal transplant outcomes: A significant independent risk factor for graft failure and patient death. Transplantation 2002; 73:70-74.

18. Yamamoto S, Hanley E, et al: The impact of obesity in renal transplantation: An analysis of paired cadaver kidneys. Clin Transplant 2002; 16:252-256.

19. Modlin CS, Flechner SM et al: Should obese patients lose weight before receiving a kidney transplant? Transplantation 1997; 64:599-604.

20. Johnson DW, Isbel NM, Brown AM, et al: The effect of obesity on renal transplant outcomes. Transplantation 2002; 74:675-681.

21. Howard RJ, Thai VB, et al: Obesity does not portent a bad outcome for kidney transplant recipients. Transplantation 2002; 73:53-55.

22. Merion RM, Twork AM et al: Obesity and renal transplantation. Surg Gynecol Obstet 1991; 172:367-376.

23. Drafts HH, Anjum MR,Wynn JJ et al impact of pre-transplant obesity on renal transplant outcomes. Clin Transplant 1997; 11:493-496.

24. Lopes IM, Martin M, Errasti P, Martinez JA: Benefits of a dietary intervention on weight loss, body composition, and lipid profile after renal transplantation. Appl Nutr Invest 1999; 15:7-10.

25 Patel MG: The effect of dietary intervention on weight gain after renal transplantation. J Ren Nutr 1998; 8:137-141.

26. Marterre WF, Hariharan S, et al:Gastric bypass in morbidly obese kidney transplant recipients. Clin Transplant 1996; 10:414-419.

27. Hricik DE, Schulak JA: Corticosteroid withdrawal after renal transplantation in the cyclosporine era: Timings, benefits and risks. BioDrugs 1997; 8:139-149.

28. Pirsch JD, D'Alessandro AM et al: Hyperlipidemia and transplantation: Etiologic factors and therapy. JAm Soc Nephro1992 (suppl 12):S238-S242.

29. Melin I, Karlsstrom B et al: A programme of behavior modification and nutrition counseling in the treatment of obesity: A randomized two year clinical trial. Int J Obes Metab Disord 2003; 27:1127-1135.

30. Coleman, DL. Effects of parabiosis of obese with diabetes and normal mice. Diabetologia 1973; 9:294.

31. Zhang, Y, Proenca et al. Positional cloning of the mouse obese gene and its human homologue. Nature 1994; 372:425.

32. Pelleymounter, MA, Cullen et al. Effects of the obese gene product on body weight regulation in ob/ob mice. Science 1995; 269:540.

33. Chen, H, Charlat, O al. Evidence that the diabetes gene encodes the leptin receptor: Identification of a mutation in the leptin receptor gene in db/ db mice. Cell 1996; 84:491.

34. Tartaglia, LA, Dembski, M et al. Identification and expression cloning of a leptin receptor, OB-R. Cell 1995; 83:1263.

35. Vaisse, C, Halaas et al. Leptin activation of Stat3 in the hypothalamus of wild-type and ob/ob mice but not db/db mice. Nat Genet 1996; 14:95.

36. Bjorbaek, C, Elmquist et al. Identification of SOCS-3 as a potential mediator of central leptin resistance. Mol Cell 1998; 1:619.

37. Chen K, Li F. et al. Induction of leptin resistance through direct interaction of C-reactive protein with leptin. Nat Med 2006; 12:425.

38. Van Heek, M, Compton, et al. Diet-induced obese mice develop peripheral, but not central, resistance to leptin. J Clin Invest 1997; 99:385.

39. Banks, WA, Kastin, et al. Leptin enters the brain by a saturable system independent of insulin. Peptides 1996; 17:305.

40. Baskin, DG, Blevins, et al. How the brain regulates food intake and body weight: the role of leptin. J Pediatr Endocrinol Metab 2001; 14 Suppl 6:1417

41. Schwartz, MW, Erickson, et al. Effect of fasting and leptin deficiency on hypothalamic neuropeptide Y gene transcription in vivo revealed by expression of a lacZ reporter gene. Endocrinology 1998; 139:2629.

42. Mizuno, TM, Kleopoulos, et al. Hypothalamic pro-opiomelanocortin mRNA is reduced by fasting and [corrected] in ob/ob and db/db mice, but is stimulated by leptin [published erratum appears in Diabetes 1998 Apr;47(4):696]. Diabetes 1998; 47:294.

43. Erickson, JC, Hollopeter, et al. Attenuation of the obesity syndrome of ob/ob mice by the loss of neuropeptide Y. Science 1996; 274:1704.

44. Yaswen, L, Diehl et al. Obesity in the mouse model of pro-opiomelanocortin deficiency responds to peripheral melanocortin. Nat Med 1999; 5:1066.

45. Bodary, PF, Westrick, et al. Effect of leptin on arterial thrombosis following vascular injury in mice. JAMA 2002; 287:1706.

46. Ducy, P, Amling, M, et al. Leptin inhibits bone formation through a hypothalamic relay: a central control of bone mass. Cell 2000; 100:197.

47. Steppan, CM, Crawford, DT, Chidsey-Frink, KL, et al. Leptin is a potent stimulator of bone growth in ob/ob mice. Regul Pept 2000; 92:73.

48. Hamrick, MW, Pennington, C, Newton, D, et al. Leptin deficiency produces contrasting phenotypes in bones of the limb and spine. Bone 2004; 34:376

48.Schett, G, Kiechl, S, Bonora, E, et al. Serum leptin level and the risk of nontraumatic fracture. Am J Med 2004; 117:952.

49. Thomas, T, Burguera, B, Melton LJ, 3rd, et al. Role of serum leptin, insulin, and estrogen levels as potential mediators of the relationship between fat mass and bone mineral density in men versus women. Bone 2001; 29:114.

50. Blain, H, Vuillemin, A, Guillemin, F, et al. Serum leptin level is a predictor of bone mineral density in postmenopausal women. J Clin Endocrinol Metab 2002; 87:1030.

51. Sato, M, Takeda, N, Sarui, H, et al. Association between serum leptin concentrations and bone mineral density, and biochemical markers of bone turnover in adult men. J Clin Endocrinol Metab 2001; 86:5273.

52. Ruhl, CE, Everhart, JE. Relationship of serum leptin concentration with bone mineral density in the United States population. J Bone Miner Res 2002; 17:1896.

53. Blum, M, Harris, SS, Must, A, et al. Leptin, body composition and bone mineral density in premenopausal women. Calcif Tissue Int 2003; 73:27.

54. Lorentzon, M, Landin, K, Mellstrom, D, Ohlsson, C. Leptin Is a Negative Independent Predictor of Areal BMD and Cortical Bone Size in Young Adult Swedish Men. J Bone Miner Res 2006; 21:1871.

55. Warren, MP, Voussoughian, F, Geer, EB, et al. Functional hypothalamic amenorrhea: hypoleptinemia and disordered eating. J Clin Endocrinol Metab 1999; 84:873.

56. Miller, KK, Parulekar, MS, Schoenfeld, E, et al. Decreased leptin levels in normal weight women with hypothalamic amenorrhea: the effects of body composition and nutritional intake. J Clin Endocrinol Metab 1998; 83:2309.

57. Harigaya, A, Nagashima, K, Nako, Y, Morikawa, A. Relationship between concentration of serum leptin and fetal growth [published erratum appears in J Clin Endocrinol Metab 1999 Mar;84(3):3106]. J Clin Endocrinol Metab 1997; 82:3281.

58. Considine, RV, Sinha, MK, Heiman, ML, et al. Serum immunoreactive-leptin concentrations in normal-weight and obese humans. N Engl J Med 1996; 334:292.

59. Saad, MF, Riad-Gabriel, MG, Khan, A, et al. Diurnal and ultradian rhythmicity of plasma leptin: effects of gender and adiposity. J Clin Endocrinol Metab 1998; 83:453

60. Kennedy, A, Gettys, TW, Watson, P, et al. The metabolic significance of leptin in humans: gender-based differences in relationship to adiposity, insulin sensitivity, and energy expenditure. J Clin Endocrinol Metab 1997; 82:1293.

61. Ostlund, RE Jr, Yang, JW, Klein, S, Gingerich, R. Relation between plasma leptin concentration and body fat, gender, diet, age, and metabolic covariates. J Clin Endocrinol Metab 1996; 81:3909.

62. Butte, NF, Hopkinson, JM, Nicolson, MA. Leptin in human reproduction: serum leptin levels in pregnant and lactating women. J Clin Endocrinol Metab 1997; 82:585.

63. Toschke, AM, Vignerova, J, Lhotska, L, et al. Overweight and obesity in 6- to 14-year-old Czech children in 1991: protective effect of breast-feeding. J Pediatr 2002; 141:764

64. Smith-Kirwin, SM, O'Connor, DM, De Johnston, J, et al. Leptin expression in human mammary epithelial cells and breast milk. J Clin Endocrinol Metab 1998; 83:1810.

65. Lyle, RE, Kincaid, SC, Bryant, JC, et al. Human milk contains detectable levels of immunoreactive leptin. Adv Exp Med Biol 2001; 501:87.

66. Matkovic, V, Ilich, JZ, Skugor, M, et al. Leptin is inversely related to age at menarche in human females. J Clin Endocrinol Metab 1997; 82:3239

67. Rosenbaum, M, Murphy EM, Heymsfield SB, et al. Low dose leptin administration reverses effects of sustained weight-reduction on energy expenditure and circulating concentrations of thyroid hormones. J Clin Endocrinol Metab 2002; 87:2391.

68. Danadian, K, Suprasongsin, C, Janosky, JE, Arslanian, S. Leptin in African-American children. J Pediatr Endocrinol Metab 1999; 12:639.

69. Haffner, SM, Stern, MP, Miettinen, H, et al. Leptin concentrations in diabetic and nondiabetic Mexican-Americans. Diabetes 1996; 45:822.

70. Boden, G, Chen, X, Kolaczynski, JW, Polansky, M. Effects of prolonged hyperinsulinemia on serum leptin in normal human subjects. J Clin Invest 1997; 100:1107.

71. Mantzoros, CS, Ozata, M, Negrao, AB, et al. Synchronicity of frequently sampled thyrotropin (TSH) and leptin concentrations in healthy adults and

leptin-deficient subjects: evidence for possible partial TSH regulation by leptin in humans. J Clin Endocrinol Metab 2001; 86:3284.

72. Schoeller, DA, Cella, LK, Sinha, MK, Caro, JF. Entrainment of the diurnal rhythm of plasma leptin to meal timing. J Clin Invest 1997; 100:1882.

73. Schorr, U, Blaschke, K, Turan, S, et al. Relationship between angiotensinogen, leptin and blood pressure levels in young normotensive men. J Hypertens 1998; 16:1475.

74. 46 Hirose, H, Saito, I, Tsujioka, M, et al. The obese gene product, leptin: possible role in obesity-related hypertension in adolescents. J Hypertens 1998; 16:2007.

75. Agata, J, Masuda, A, Takada, M, et al. High plasma immunoreactive leptin level in essential hypertension. Am J Hypertens 1997; 10:1171.

76. Weigle, DS, Duell, PB, Connor, WE, et al. Effect of fasting, refeeding, and dietary fat restriction on plasma leptin levels. J Clin Endocrinol Metab 1997; 82:561.

77. Farooqi, IS, Matarese G, Lord BM. Beneficial effects of leptin on obesity. T cell hyporesponsivenss, and neuroendocrine/metabolic dysfunction of human congenital leptin deficiency. J Clin Invest 2002; 110:1093.

78. Ozata, M, Ozdemir, IC, Licinio, J. Human leptin deficiency caused by a missense mutation: multiple endocrine defects, decreased sympathetic tone, and immune system dysfunction indicate new targets for leptin action, greater central than peripheral resistance to the effects of leptin, and spontaneous correction of leptin-mediated defects. J Clin Endocrinol Metab 1999; 84:3686.

79. Farooqi, IS, Jebb, SA, Langmack, G, et al. Effects of recombinant leptin therapy in a child with congenital leptin deficiency. N Engl J Med 1999; 341:879.

80. Farooqi, IS, Keogh, JM, Kamath, S, et al. Partial leptin deficiency and human adiposity. Nature 2001; 414:34

81. Licinio, J, Caglayan, S, Ozata, M, et al. Phenotypic effects of leptin replacement on morbid obesity, diabetes mellitus, hypogonadism, and behavior in leptin-deficient adults. Proc Natl Acad Sci U S A 2004; 101:4531.

82. Clement, K, Vaisse, C, Lahlou, N, et al. A mutation in the huam leptin receptor gene causes obesity and pituitary dysfunction. Nature 1998; 392:398.

83. Farooqi, IS, Wangensteen, T, Collins, S, et al. Clinical and molecular genetic spectrum of congenital deficiency of the leptin receptor. N Engl J Med 2007; 356:237.

84. Considine, RV, Considine, EL, Williams, CJ, et al. Evidence against either a premature stop codon or the absence of obese gene mRNA in human obesity. J Clin Invest 1995; 95:2986.

85. Maffei, M, Stoffel, M, Barone, M, et al. Absence of mutations in the human OB gene in obese/diabetic subjects. Diabetes 1996; 45:679.

86. Heymsfield, SB, Greenberg, AS, Fujioka, K, et al. Recombinant leptin for weight loss in obese and lean adults: a randomized, controlled, dose-escalation trial. JAMA 1999; 282:1568.

87. Welt, CK, Chan, JL, Bullen, J, et al. Recombinant human leptin in women with hypothalamic amenorrhea. N Engl J Med 2004; 351:987.

88. Chan, JL, Mantzoros, CS. Role of leptin in energy-deprivation states: normal human physiology and clinical implications for hypothalamic amenorrhoea and anorexia nervosa. Lancet 2005; 366:74.

89. F. Cof 'ana, E. Velab et al. Obesity in Renal Transplantation: Analysis of 2691 Patients. Transplantation Proceedings.2005:3695-3697.

90. Kooman, Eugénie C. H. et al. Post Transplantation weight gain is predominantly due to an increase in body fat mass. 2000: july 15, 70:1

91. Hart PD, Wilkie ME et al. Dual energy X-ray absorptiometry versus skinfold measurements in the assessment of total body fat in renal transplant recipients. Eur J Clin Nutr. 1993 May; 47(5); 347-52.

92. Alfonso M et al. Bone loss in long-term renal transplantation: Histopathology and densitometry analysis. Kidney international, 1999; 55:2021-2079.

93. Fabio. M. Ulivieri et al. Effect of kidney transplantation n bone mass and ody composition in Males. Transplantation. 2002; 73:612-615.

94. Horber FF, Casez JP et al. Changes in bone mass early after kidney transplantation. J Bone Miner Res. 1994; 9:1.

95. Kwan JT, Almond MK et al. Changes in total body bone mineral content and regional bone mineral density in renal patients following renal transplantation. Miner Electrolyte Metab. 1992;18:166.

96. Cuteo-Manzano AM, Konel S et al. Bone loss in long term renal transplantation: Histopathology and densitometry analysis. Kidney Int. 1999; 55:2021-2129.

97. Mathieu RL, Casez JP et al. Altered body composition and fuel metabolism in stable kidney transplant patients on immunosuppressive monotherapy with cyclosporine. A. Eur J Clin Invest. 1994;24:195.

98. Steiger U, Lippuner K et al. Body composition and fuel metabolism after kidney grafting. Eur J Clin Invest.1995;25:809.

99. Van den Ham EC, Kooman JP et al. Relation between steroid dose, body compostion and physical activity in renal transplant patients. Transplantation. 2000;69:1591.

100. Ostlund RE jr. Yang IW et al. Relation between plasma leptin concentratin and body fat, gender, diet, age and metabolic co-variates. J.Clin. Endocrinol Met. 1996;81:3909-13.

101. Agras PI, Bastin E et al. Relationship between leptin and bone mineral density in renal transplant recipients. Transplant Proc.2005; 37(7):3106-8.

102. Malyaszko J, Malyszko JS. Et al. Correlation between leptin, body composition, bone mineral density and bone metabolism in kidney transplant recipients. Transplant Proc. 2005; 37(5):2151-3

103. Franciszek Kokot, Marcin Adamczak et al. Plasma leptin concentration in kidney transplant patients during early post transplantation period. Nephrol Dial Transplant. 19998; 13:2276-80.

Proforma

Date:
SL No:

Name:
Age:
Sex: M/F
Hosp No:
Height:

	Weight (kg)	Leptin (ng/ml)
Baseline		
3 months		
6 months		

Caliper Measurements:

(mm)	Baseline	3 months	6 months
Biceps			
Triceps			
Subscapular			
Suprailiac			
Total			

DEXA measurements:

	Bone mineral content			Bone mineral density		
	Baseline	3 months	6 months	Baseline	3 months	6 months
Head						
Arm						
Rib						
Trunk						
Spine						
Leg						
Pelvis						
Total						

	Fat (gms)			Fat (%)			Lean (gms)		
	B/L	3 mo	6 mo	B/L	3 mo	6 mo	B/L	3 mo	6 mo
Head									
Arm									
Trunk									
Leg									
Total									